U.S. DAIRY FORAGE RESEARCH CENTER

1989 RESEARCH SUMMARIES

U.S. Dairy Forage Research Center
1925 Linden Drive West
Madison, Wisconsin 53706

Agricultural Research Service

United States Department of Agriculture

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U.S. DAIRY FORAGE RESEARCH CENTER, USDA-ARS
Madison, WI 53706

Dear Reader:

It is a pleasure to update our progress by bringing you these summaries of recent research. The U.S. Dairy Forage Research Center is a unique part of the national research program of the Agricultural Research Service, U.S. Department of Agriculture. The Center's mission is to build a knowledge and technology base for the dairy industry to fully exploit the use of forages in the production of milk. The Center has agricultural engineers, plant and soil scientists, microbiologists, ruminant nutritionists and a chemist working together to increase the efficiency of forage production and utilization by dairy farmers. We function in close cooperation with the Agricultural Experiment Stations of several states. The Center is located on the campus of the University of Wisconsin, Madison, and has "Cluster" locations in St. Paul, MN, Ames, IA, Columbia, MO, Wooster, OH, East Lansing, MI, University Park, PA and Ithaca, NY. The Center's research farm, with facilities for 300 milking cows, is located on 63 acres of USDA land on the banks of the Wisconsin River in Prairie du Sac, WI. An additional 1200 acres of adjacent land is utilized by the Center by agreement with the U.S. Department of the Army.

The Center was established in 1980 and has made steady growth since. At present there are eighteen scientists; ten at Madison, and one at each of six Cluster locations, and two at the St. Paul, Minnesota Cluster location. Scientists hold faculty appointments in university departments and provide supervision for approximately 20-25 graduate students and 4-8 post doctoral fellows.

The Center is making a special effort to obtain Nuclear Magnetic Resonance (NMR) instrumentation this year. This will be crucial for the continued development of our understanding of the forage cell wall. Significant groundwork is being laid in the area of cell wall chemistry, and we are optimistic about the progress that is achievable in the next few years. The Cell Wall Group within the USDFRC is currently planning for an International Symposium on the forage cell wall in October, 1991. This could be an important stimulus for work in that area.

Environmental issues will occupy center stage of U.S. agriculture in the 1990's. Forages will play an important role in cropping plans designed to minimize soil loss and maintain water quality. Because of heavy reliance on forages, dairy farmers as a whole have been good stewards of our soil and water resources. However, the dairy industry may be living closer to the line than is comfortable regarding potential ground water contamination with nitrate. The typical dairy farm, growing nitrogen fixing legume forages and purchasing considerable amounts of protein supplement, adds a very large amount of nitrogen into the system. If good manure management is not followed, nitrate leaching into the ground water is possible. It is unfortunate that the protein in legume forages is so easily degraded in the cow's rumen. This requires purchase of protein supplements in larger amounts then would otherwise be needed. We need to find ways of modifying the protein in forages, either genetically or by post-harvest treatment, to improve the efficiency of protein utilization by the cow. This will not only reduce costs, but could reduce total nitrogen input into the cycle, thus reducing potential for nitrate contamination of ground water.

We are pleased and very proud of the way Center scientists from diverse disciplines interact and bring their collective insights to bear on the problems of forage production and utilization. This collection of research summaries illustrates the progress they are making in developing information to help dairy farmers utilize their resources more effectively. The research is intended to benefit dairy farmers and the consumers of dairy products.

Sincerely,

Larry D. Satter

Larry D. Satter, Director
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Acknowledgments

Appreciation is expressed to Gloria Palmer for her interest in and dedication to the task of typing and assembling this annual research summary.

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RESEARCH PROCEDURES

CALCULATING THE IMPACT OF ALFALFA ON THE NITROGEN CYCLE IN THE NORTH CENTRAL REGION

T.A. PETERSON and M.P. RUSSELLE

Public concern about the environment has prompted examination of agricultural nitrogen (N) cycles. Accumulating evidence shows that some production practices may contribute N to ground water. Crop rotations involving legumes have been suggested as a way to reduce fertilizer N applications, thereby decreasing N losses, and reducing leaching of NO_3-N to ground water. Our objective was to analyze the impact of alfalfa on inputs and potential losses of N in the North Central USA. Specifically we examined the inputs of symbiotically-fixed nitrogen (SFN) from alfalfa to the soil, both from direct transfer and from feed N recycled as manure. We calculated potential savings of fertilzer N applied to corn assuming proper credit was given to manure applications and preceding alfalfa crops.

About 40% of US alfalfa hay production comes from the North Central Region (IL, IN, IA, MI, MN, MO, OH, and WI). Annual dry hay production in these 8 states averaged 33 million Mg (metric ton) from 1984-1986 (Table 1), with an additional 4 million Mg hay equivalent harvested as haylage or green chop (assuming 85% of haylage is taken from alfalfa fields). Wisconsin produces 30% of the region's alfalfa. Our estimate of alfalfa production is low as data are not available for extent and productivity of alfalfa pastures. For this analysis, we assumed alfalfa is rotated to corn at 3-yr intervals, that production in each state was evenly divided between 1st-, 2nd-, and 3rd-year stands, and that there was no net import or export of alfalfa for the region. We assumed alfalfa herbage contains 2.8% N and that 50% of herbage N came from symbiotic fixation the first year, with 80% of herbage N derived from fixation in years 2 and 3. About 640 million kg of SFN are harvested annually in alfalfa forage in the North Central Region (Table 1). Annual removal of N from "soil" (i.e., sources other than symbiotic fixation) averaged 58 kg/ha.

Alfalfa contributes about 56 kg N/ha to the soil annually. Assuming this increase occurs in addition to the amount of soil N removed in herbage, we calculate the annual fixed N_2 input to soil is about 475 million kg (or 114 kg N/ha) for the region. Thus, annual SFN from alfalfa totals over 1.1 billion kg in the 8-state region. Annual sales of N fertilizer in the region averaged 4 billion kg, with over 3 billion of that total applied to corn. Therefore, although alfalfa was grown on only 8% of the land area in the North Central Region, SFN from alfalfa represents a substantial and concentrated input to the agricultural N cycle.

There are many uncertainties in estimating the fate of SFN when alfalfa is fed to animals. We simplified the system to include only two classes of animals, milking cows and others. We calculated the amount of SFN in livestock products by assuming one-half of the N fed to milking cows comes from alfalfa, that milk contains 3.2% protein, and that the feed-to-milk N conversion factor is 0.3. We used a conversion factor of 0.15 for the "other" animal products. Ammonia loss estimates from manure are quite variable; we used estimates from Van Dyne and Gilbertson (1979). Ammonia losses total 39% of the SFN in manure, or 32% of the SFN originally in feed.

Few producers properly reduce fertilizer N rates applied to corn following a legume or manure application. We calculated the reduction in fertilizer N application possible if proper credit is given for manure and previous alfalfa crop. If these recommendations are followed, fertilizer N use in the North Central Region would be reduced by up to 18%.

Table 1. Impact of alfalfa on the agricultural N cycle, return of SFN to soil in manure, and potential reductions in fertilizer N applied to corn in Wisconsin and the North Central Region.

	Wisconsin	N.C. Region	Units
A. Alfalfa Forage Production			
Alfalfa area	1269	4168	10^3 ha
Dry hay (15% moisture)	9.20	33.00	10^6 Mg
Haylage (Haylage:dry hay, 3:1)	1.92	4.08	"
Total	11.21	37.08	"
Nitrogen in forage			
Atmospheric N	194	641	10^6 kg
Soil N (i.e., all other sources)	73	241	"
Total	267	882	"
Soil Removal + N Buildup (buildup= 56 kg/ha)	144	475	"
Total N_2 fixation	338	1116	"
Fertilizer N applied to corn	173	3089	"
B. Disposition of Symbiotically Fixed N_2			
SFN Fed to milking cows (0.5 of dietary N)	70	164	"
SFN Fed to other livestock	124	477	"
Total SFN fed in forage	194	641	"
SFN in manure			
Milking cows (SFN in feed x 0.7)	49	115	"
Other (SFN in feed x 0.85)	105	405	"
Total	154	520	"
Ammonia losses (25% for dairy and 30% for other during storage + 15% loss during application)	60	205	"
C. Potential Reductions in Fertilizer N Applied to Corn			
Corn area after alfalfa (1/3 alfalfa area)	423	1389	10^3 ha
Potential reductions after alfalfa			
0 N is applied in yr 1, 50% N rate in yr 2	67	277	10^6 kg
50% N rate applied in yr 1, 100% N rate in yr 2	33	92	"
Potential reductions with manure applications			
40% of the manure N available in yr 1, and 20% of remainder available in yr 2	80	270	"
30% of the manure N available in yr 1, and 15% of remainder available in yr 2	62	211	"
Estimated range of fertilizer N reductions:			
Sum of low estimates	96	303	"
Sum of high estimates	147	548	"

EFFECTS OF SEEDING YEAR SUMMER HARVEST SCHEDULES ON ALFALFA YIELDS

K.A. TOELKE and R.P. WALGENBACH

Introduction

The practice of establishing alfalfa with a companion crop has resulted in conservative harvesting in the seeding year. The increasing acceptance of direct seeding and the availability of herbicides for weed control creates a need for management strategies which will optimize yield and quality without compromising future productivity. Alfalfa cultivars with improved vigor, disease resistance and good regrowth potential should allow more intensive management in the seeding year with good winter survival. The objectives of this study were to determine the influence of alfalfa cultivars and seeding year summer and autumn cutting schedules on alfalfa yield, chemical composition, stand persistence and productivity. This progress report will discuss seeding year summer cutting schedule effects on yield. A future report will discuss summer and autumn cutting schedule effects on productivity and persistence.

Materials and Methods

Field plots of Blazer, WL225 and Legand Alfalfa were seeded on approximately 20 April 1988 and 1989 in a randomized complete block design in a split-split plot arrangement. The whole plots were summer harvest schedules beginning approximately 50, 60 and 75 days post emergence with subsequent summer harvests at 30 or 40 day intervals (Table 1). Three autumn cutting schedules were superimposed on each summer schedule. Cultivars comprised sub-sub plots. In 1988 plots received 7 cm of irrigation to aid establishment. Yield data for autumn harvests are not reported here and forage quality analysis is incomplete at this writing.

Results and Discussion

In both seeding years, field plots were stressed by drought conditions, resulting in reduced yields. Higher first harvest yields in 1988 compared to 1989 are probably due to early season irrigation and planting in soil that was previously fallow. First harvest yields increased as the harvest date was delayed after emergence (Table II). While 1794 kg of dry matter had accumulated in 50 days after seeding (Schedule I, Table I), that amount was nearly doubled by delaying harvesting for 25 days (Schedule L) in 1988. Similar results were seen in 1989. Schedule I was cut at mid vegetative stage and would provide very high quality forage. In 1988, harvest schedule M produced the largest amount of forage (Table II).

All plots were uniform and appeared fairly vigorous going into the autumn despite dry weather conditions. Plants in schedule L regrew more rapidly from the 17 August cutting than did plants in Schedule I which was cut only two days later in both 1988 and 1989. We think that a wide range of options exist for seeding year summer cutting of alfalfa which should allow for flexible cutting schedules.

Table 1. Summer cutting dates for the 1988 seeding year harvest schedules.[†]

Cutting Schedule	# Days After Emergence[‡]	Summer Harvest Dates 1st	2nd	3rd
Intensive (I)	51	6-20	7-19	8-17
Moderate (M)	61	6-30	8-1	9-1
Lenient (L)	73	7-12	8-19	—

[†]Dates were the same or 1-2 days later in 1989.

[‡]The number of days between emergence and first harvest.

Table 2. Influence of harvest date on seeding year summer yields in 1988 and 1989.

Cutting	1st harvest		2nd harvest		3rd harvest		Total	
Schedule	1988	1989	1988	1989	1988	1989	1988	1989
	kg ha^{-1}							
Intensive	1794	1280	2383	1242	2350	2435	6527	4957
Moderate	2666	1933	2955	2004	2052	2095	7673	6032
Lenient	3497	2461	2981	3474	—	—	6478	5936
LDS .05	135	142	439	316	268	212	891	473

SELECTION FOR RESISTANCE TO *APHANOMYCES EUTEICHES* IN RED CLOVER

J.E. TOFTE, R.R. SMITH and C.R. GRAU

Introduction

The pathogen *Aphanomyces euteiches* is primarily known for its yield loss capabilities in the field pea. However, it is also ascribed to be a major cause of seeding failures in alfalfa in wet, acid soils. While red clover was known to be a moderate host to the pathogen, it was not until recently that a virulent strain of *A. euteiches* (Ae572) to red clover had been isolated. Red clover seeds germinating in wet, acid soils are extremely susceptible to this pathogen. The purpose of this paper is to report on the effectiveness of selection for resistance to *A. euteiches*, strain Ae572.

Materials and Methods

Initial investigations suggested that the frequency of phenotypic resistant plants was very low. Using previously developed growth chamber screening methods, phenotypic mass selection for resistance was not extremely effective after one cycle. The disease symptoms are not expressed if the plants are over 10 days old when exposed to the pathogen. Exposing germinating seeds to the pathogen identifies many susceptible genotypes, however some seeds escape infection and are perhaps not resistant. Since symptoms are not expressed on older plants, repeat inoculation is not effective in identifying these escapes. Therefore, it appeared that perhaps progeny testing and repeated screening of progeny families would assist in identifying the most resistant plants in the most resistant families. So after the first cycle of phenotypic mass selection two cycles of progeny selection were applied to the cycle 1 material. The initial germplasm was a synthetic composed of plant material previously selected for persistence in wet, acid soils.

Results and Discussion

The results of the three cycles of selection for resistance to *A. euteiches* in red clover are presented in Table 1. As mentioned above, no significant improvement was made in the first cycle of selection (mean DSI of C0 = 3.64 vs mean DSI of C1 = 3.60). However, once progeny testing was applied after the first cycle significant progress was achieved (mean DSI of C2 = 2.81 and mean DSI of C3 = 2.43). Also note significant shift in frequency of resistant plants (DSI of 1 and 2) between cycle 1 and cycle 3. It would appear from the extremely high estimates of heritability (h^2) of 85 to 92% that selection should be very effective and that a high level of resistance would be achieved quite rapidly. On the contrary, the inability to positively identify resistant plants in the initial screening slows the actual progress and a certain frequency of susceptible plants are misclassified each cycle, thus preventing a rapid accumulation of resistant plants in the population.

Table 1. Response to selection for resistance to *Aphanomyces euteiches* in red clover.

Cycle	Percent of plants wth DSI· of 1 and 2	3	4 and 5	Mean DSI	h^2
0	2	47	51	3.64a··	92
1	5	47	48	3.60a	—
2	31	45	24	2.81b	90
3	49	34	17	3.71c	85

·DSI = Disease Severity Index: 1 = resistance, no symptoms, 5 = dead seedlings.

··Means followed by the same letter are not significantly different at the 5% level.

h^2 = heritability (%).

CHARACTERIZATION OF COMPOSITIONAL DIFFERENCES AMONG FORAGES BY PYROLYSIS/GAS CHROMATOGRAPHY

S.M. ABRAMS

Introduction

Fiber generally represents greater than 50 percent of the dry matter of forages, consisting of various complex polymers. The chemical constitution of these polymers and the way in which they are bound together govern the ultimate utilization of forages by ruminants. Spectrophotometric studies utilizing near infrared reflectance have clearly demonstrated the limits of many conventional extractive techniques in understanding these relationships.

Pyrolysis is the process of molecular fragmentation of polymers which occurs when they are exposed to intense heat in the absence of oxygen. These fragments, being volatile, can be separated and quantified by gas chromatography. Recent Dutch work in wood chemistry suggests that these fragments can be related back to their starting polymers. Because the nature of the end products and the pattern of fragmentation is peculiar to the starting material, pyrograms represent characteristic fingerprints of the original material; the technique is often used in forensic work to make positive identification of materials such as hair, cloth, etc. Pyrolysis products (pyrolyzates) may be quantitatively related to utilization of forages by ruminants.

The objectives of this study were to develop a system for pyrolysis/GC analysis of forages, and ultimately to characterize differences between forages known to vary in quality, as defined by either animal utilization or in vitro procedures.

Materials and Methods

The analysis system consists of a CDS 120 pyrolysis unit, interfaced with an HP 5890 Gas Chromatograph. Data are processed by an HP 3396A integrator and collected on a Zenith 248 computer. Approximately 150 micrograms of sample are pyrolyzed under helium for 10 seconds at 650°C in a heated (225°C) interface. Volatiles are swept into the injector port of the GC, which is fitted with a 30 meter capillary column (DB1701). Oven temperature is maintained at 35°C for 2 minutes, raised to 165°C at 3°C per minute, raised to 270°C at 6°C per minute, and held at 270°C for 2 minutes. The split ratio is maintained at 100:1.

Results

Both pure materials (cellulose, glucose, xylose) and forages have been examined. Sample pyrograms of three hays are presented in Figure 1. Pyrograms of both alfalfas are very similar as to the location of prominent peaks, with principal variation consisting mainly in small differences in the magnitude of the different pyrolysates. The orchardgrass pyrogram show greater differences, although prominent peaks have similar retention times for all three forages. Peaks prominent in cellulose and glucose pyrograms (not shown), with retention times of 3.9 and 51.4 minutes, are present in all three forages. Duplicate analyses on the same sample have shown good agreement. These preliminary findings demonstrate that in spite of the small sample sizes used, pyrograms can be produced that are characteristic of the forage of interest.

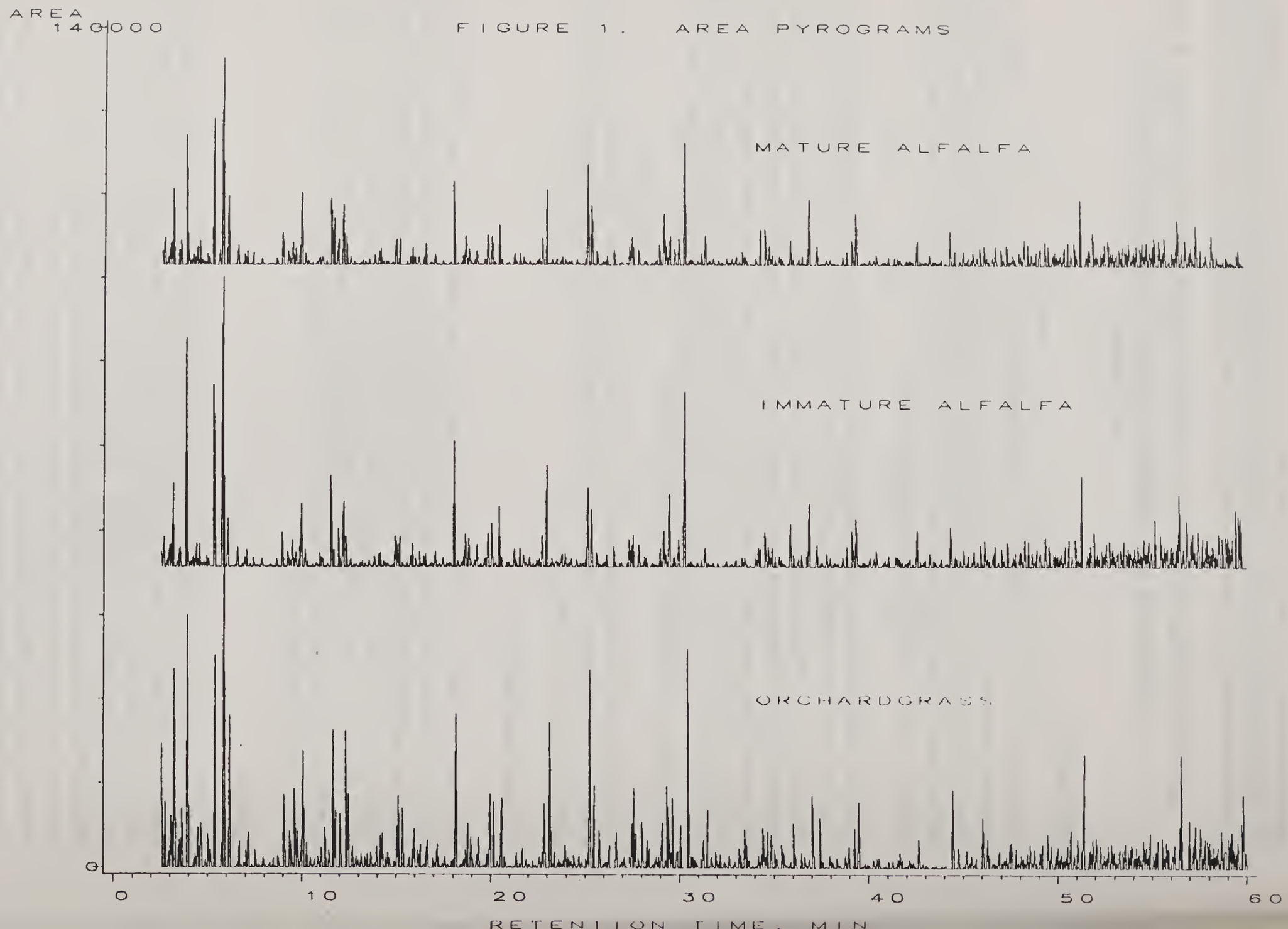


FIGURE 1. AREA PYROGRAMS

PYROLYSIS-GC-MS FOR THE CHARACTERIZATION OF FORAGE MATERIALS

J. RALPH and R.D. HATFIELD

Introduction

Plant materials have been shown to pyrolyse, in the absence of air at temperatures up to 800°C, to give fragments which are diagnostic of their parent polymers. The volatile fragment compounds can be separated by gas chromatography (GC) and they can often be identified by mass spectrometry (MS). Consequently, the hybrid technique of pyrolysis-GC-MS is a very useful method for obtaining useful data from relatively small amounts of material and in a relatively short time.

The Technique

Pyrolysis was carried out using a CDS pyroprobe set to ballistically (>50°C/ms) heat samples of 250 to 750 μg of plant materials to 700°C and to carry the volatile components onto the top of a DB-1 capillary column in an HP 5890 GC. The GC temperature was held at 50°C for 2 minutes to trap and focus the volatile materials and was then ramped to 275°C at 4°C/min. On the 60 m column used, compounds of interest eluted between 20 and 60 minutes. These comprised compounds readily identifiable by their mass spectra (HP5870 mass selective detector with Unix datastation) as lignin-related, carbohydrate-related, or extractives.

Lignin fragments are trivial to elucidate structurally due to their logical and diagnostic fragmentation pathways in mass spectrometry. Carbohydrate-derived compounds are much harder but we are helped by the excellent work of other researchers in the field.

A typical pyrogram and some of the identifiable compounds are shown in Figures 1 and 2. A mass spectrum of 4-vinyl-guaiacol, a compound resulting largely from ferulic acid in the tissue, is shown in Figure 3. The elucidation of structures for other peaks is left as an exercise to the reader.

Applications and Value

We are still somewhat in the exploratory stages of this technique but it looks to be an excellent screening method for various cell wall components, particularly lignins and the phenolic acids which fragment very diagnostically. It also provides an excellent qualitative way of gaining an overview of differences in cell tissues from plants at various stages of development, from different parts of a plant, and from plant cell wall material that has been subjected to various treatments, including digestion by ruminants. Now that this labour-intensive ground work of identifying major pyrogram peaks is essentially complete, we can look forward to exciting applications in many areas of our research dealing with digestion and processing of plant materials.

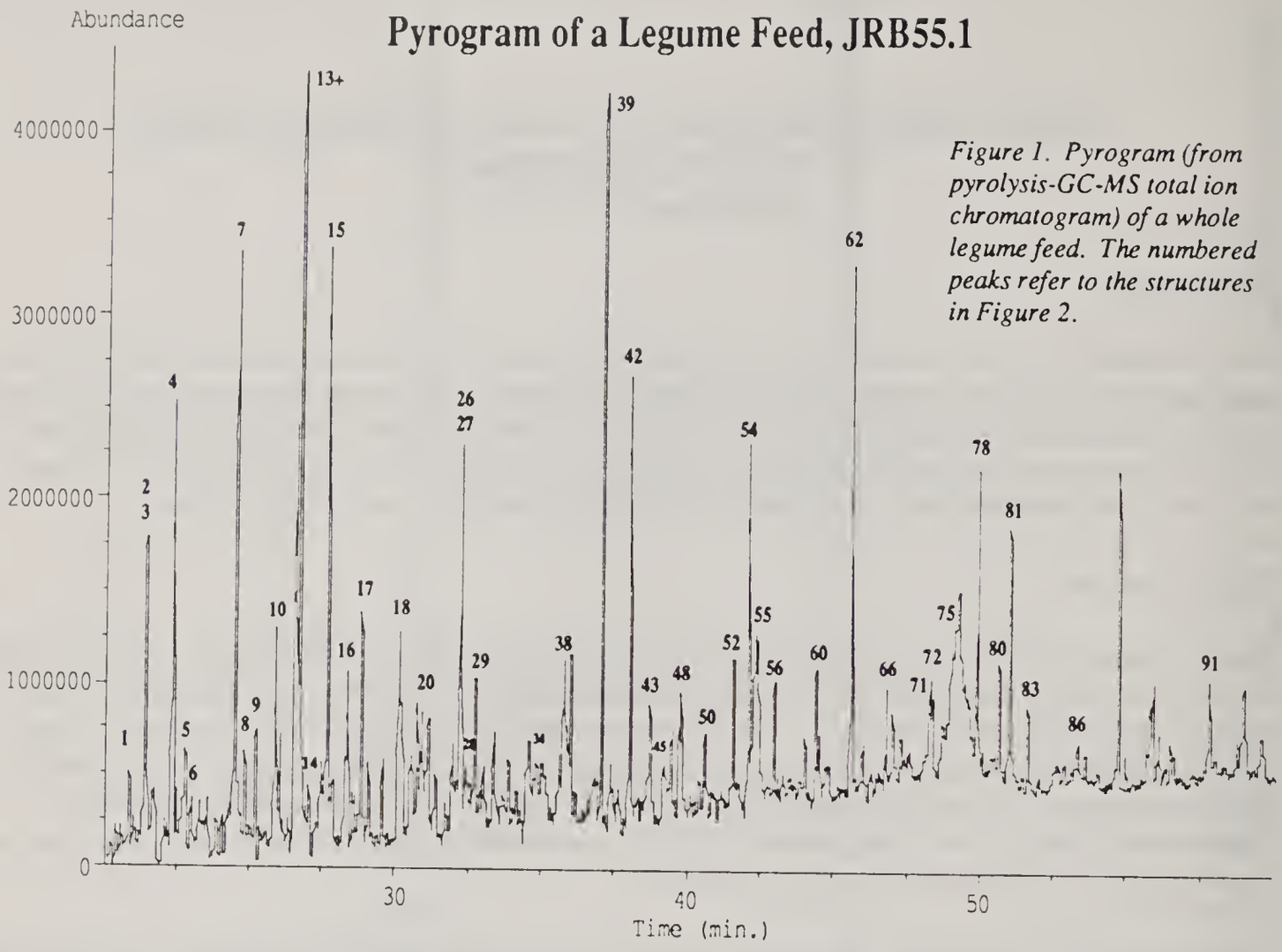


Figure 1. Pyrogram (from pyrolysis-GC-MS total ion chromatogram) of a whole legume feed. The numbered peaks refer to the structures in Figure 2.

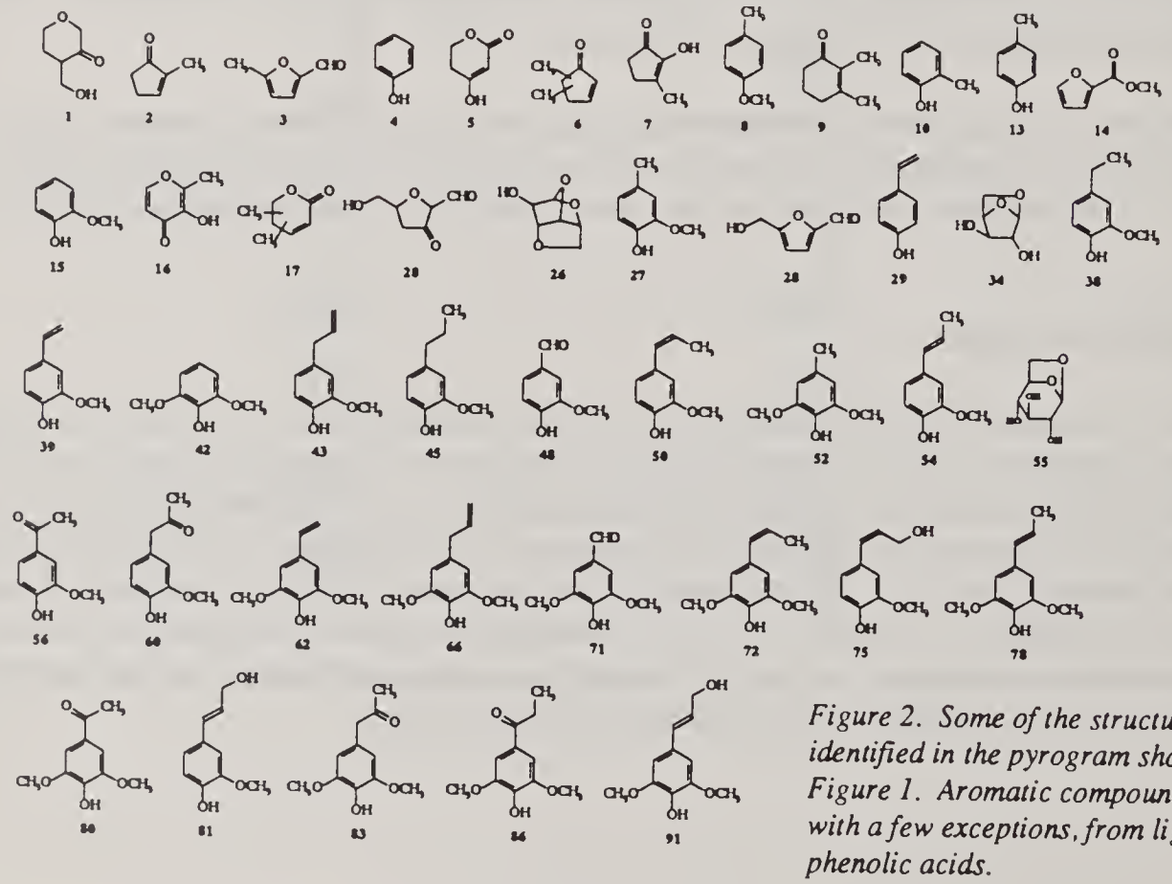


Figure 2. Some of the structures identified in the pyrogram show in Figure 1. Aromatic compounds arise, with a few exceptions, from lignin and phenolic acids.

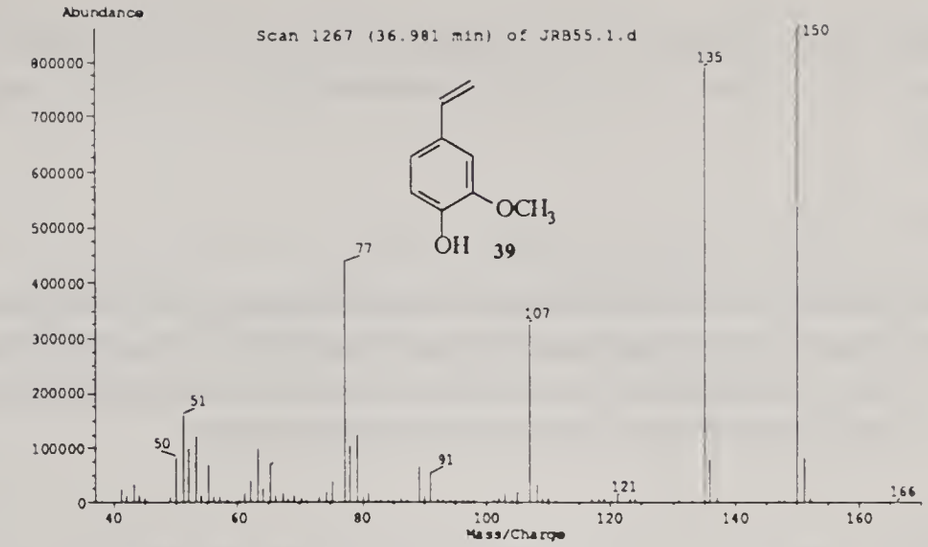


Figure 3. The mass spectrum of compound 39, vinyl guaiacol, which arises substantially from ferulic acid derivatives in the cell wall. The molecular weight of vinyl guaiacol is 150.

SYNTHESIS OF COMPOUNDS WHICH MODEL LIGNIN-PHENOLIC ACID STRUCTURES IN THE PLANT CELL WALL

J. RALPH and R. HELM

Introduction

Phenolic acids may play an important role in limiting the digestibility of plant cell walls because of their ability to cross-link cell wall polymers. Although it has been reasonably well demonstrated that phenolic acids are linked to lignin, as both esters and ethers, there is little more than speculation on the sites of attachment to the lignin polymer. Knowing the attachment sites will tell us a great deal about the processes by which phenolic acids are attached to cell wall polymers and how they are implicated in cell wall biosynthesis.

In attempting to look at the details of phenolic acids in the cell walls of plants (another story!) we first need to prepare model compounds for all of the most probable structures. This report focuses only on those compounds which model lignin-phenolic acid structures. Current work is also aimed at synthesizing good carbohydrate-phenolic acid model compounds.

Syntheses

All compounds modelling phenolic acids attached to a ß-aryl ether unit in lignin are potentially derivable from the single lignin model compound **1**, guaiacylglycerol-ß-guaiacyl ether, although this is quite a synthetic oversimplification because of the need to juggle protecting groups. A conceptual scheme for preparing all three esters and one of the aryl ethers is shown in Figure 1. The phenolic- and the γ-ester are potentially available from normal esterification reactions, while the α-ester and the α-ether can be obtained by reactions of the quinone methide **2** derived from **1**. The ß-ether needs to be synthesized quite separately as outlined in Figure 2.

The syntheses of all of these compounds, for both ferulic acid and *p*-coumaric acid is now complete. They still need to be fully characterized by mass spectrometry and will be subjected to very rigorous structural and conformational analysis by a plethora of modern NMR techniques and by 3D molecular modelling.

Applications and Value

Some of these models have a value in their own right and are being sought by other researchers in related fields. However, their real value derives from their spectroscopic information. Well chosen model compounds will give us the data we need to find and determine the detailed structure of these components in the plant cell wall. That work is about to begin.

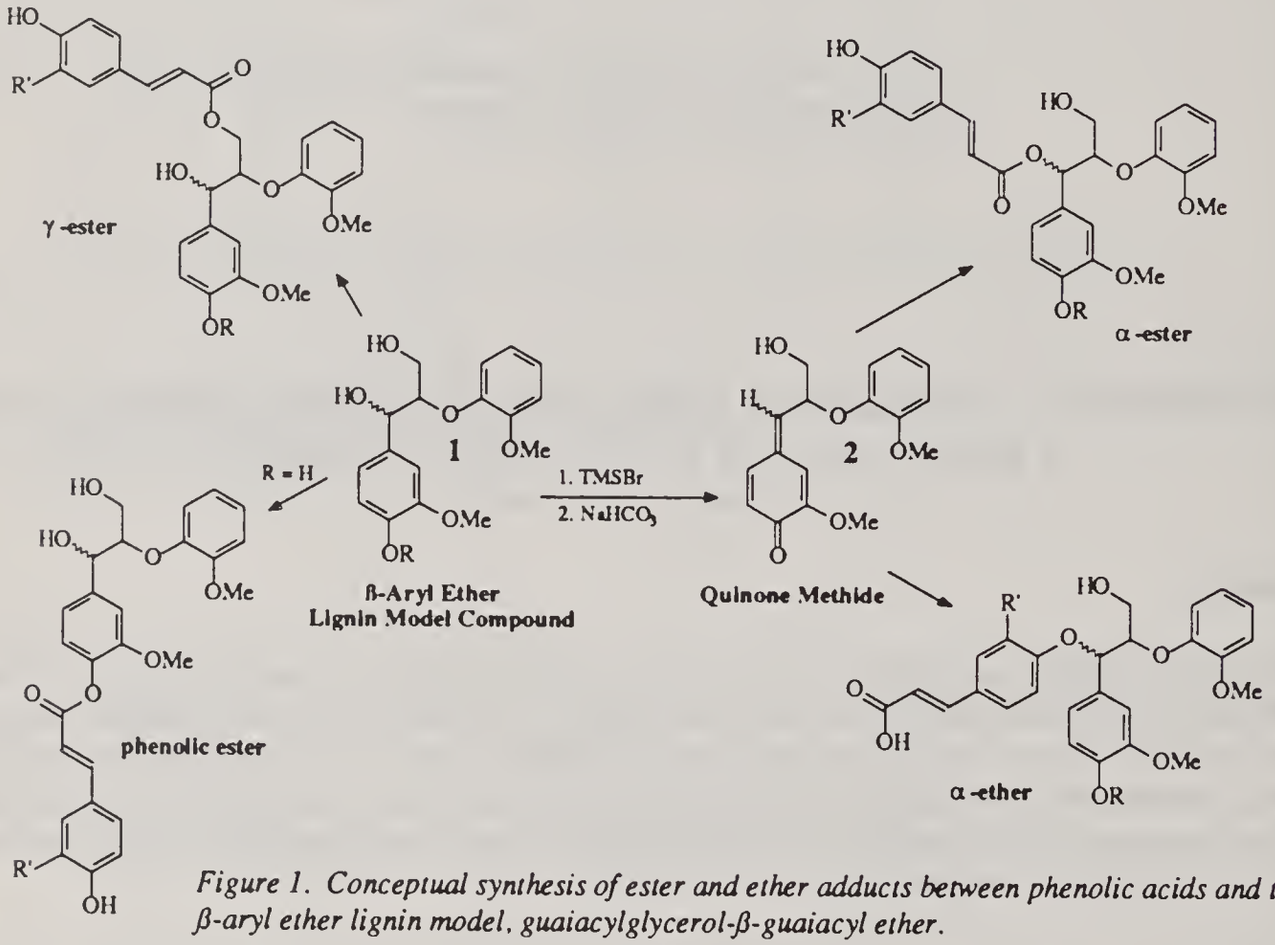


Figure 1. Conceptual synthesis of ester and ether adducts between phenolic acids and the ß-aryl ether lignin model, guaiacylglycerol-ß-guaiacyl ether.

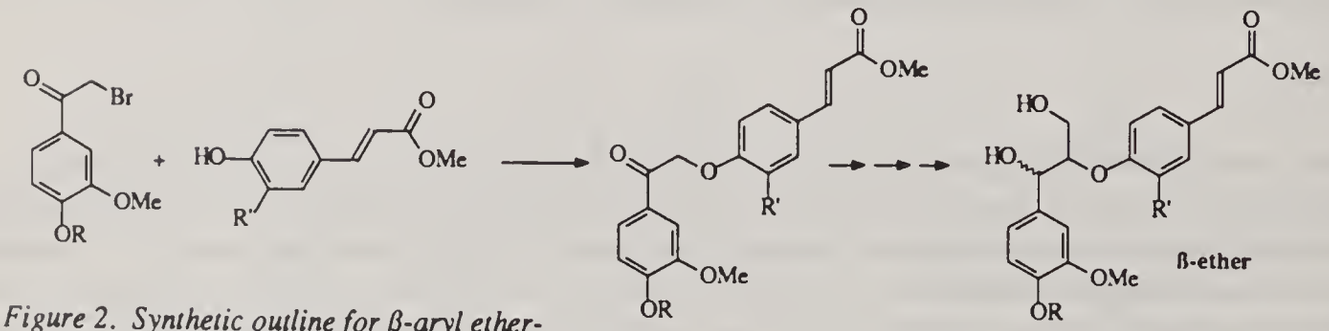


Figure 2. Synthetic outline for ß-aryl ether-phenolic acid compounds.

DETAILED STRUCTURAL ANALYSIS OF ALFALFA PECTINS

R.D. HATFIELD

Introduction

Alfalfa contains significant amounts of pectic materials. Their degradation pattern by microorganisms is not known. Characterizing the physio-chemical properties of these polysaccharides will lead to a better understanding of their role in the developing plants and their impact on cell wall utilization. We report here on the characteristics of one subclass of these molecules.

Materials and Methods

Alfalfa plants were grown in a greenhouse under high pressure sodium lamps with a 14/10 day/night regime. Plants were harvested at the early bud stage and cell walls isolated from stem sections. The milled-cell walls were suspended in phosphate buffer and heated in a boiling water bath to extract hot water soluble polysaccharides (HW). Extracted polysaccharides were fractionated by anion exchange chromatography on a DEAE Spectra/Gel M column (2.5 X 15 cm).

Pooled fractions containing the highest amounts of uronic acid collected from the DEAE column were treated with purified endopolygalacturonase. The polysaccharide and enzyme mixture were incubated for 20h at 25°C. On separate samples the methyl esters of the uronosyls were removed by treatment with 0.5 M NaOH at room temperature. The sample was cooled on ice and the pH adjusted to 5.0 with glacial acetic. Insoluble material was pelleted by centrifuging and the supernatant was carefully removed and treated with endopolygalacturonase. The insoluble material was dissolved in dH_20, made 10mM acetate with concentrated buffer and treated with the enzyme. The degree of enzyme degradation was monitored by HW-55 column chromatography. Degradation products were analyzed for neutral sugar composition and linkage patterns.

Results and Discussion

Polysaccharides in the HW extract were fractionated into neutral and charged fractions using DEAE anion exchange chromatography. Small amounts of carbohydrate did not bind to the column and eluted in the column void. Sugar composition and linkage analysis indicated that this fraction was predominantly arabinan. The majority of the polysaccharides bound to the column and eluted as a narrow peak (F4) which was approximately 56% uronosyl residues of which 41% contained methyl esters. Smaller amounts of material bound to the column and eluted just ahead of this fraction. These small peaks contained arabinose, galactose, xylose, galacturonic acid and glucuronic acid.

The F4 DEAE peak contained rhamnose (15%), galactose (24%) and arabinose (59%) as the major neutral sugars. Galacturonic acid was the only uronosyl residue detected. Undegraded F4 polysaccharides eluted rapidly from an HW-55 column with K_{av}^{1}=0.09-0.25. After treatment with endopolygalacturonase the elution profile did not change. Deesterification of this fraction resulted in the formation of a soluble (F4-S) and an insoluble (F4-I) fraction. Treatment of the soluble fraction (F4-S) with enzyme resulted in a shift to lower molecular weight fragments. However, a significant portion of the neutral sugars (70-75%) remained as polysaccharides (K_{av}=0.19-.43) with small amounts of uronic acid residues. Uronic acid enriched fractions were small molecular fragments being retained on the column longer (K_{av}=0.82-0.97).

Composition of the polysaccharide fraction (K_{av}=0.19-.43) was predominantly arabinose (54%), galactose (23%), and rhamnose (13%). Linkage analysis revealed patterns similar to arabinan and arabinogalactans. The amount of rhamnose was greater than the total uronosyl residues and contained branch sugars. Composition of the uronic rich fractions (K_{av}=0.82-0.97) was predominantly galacturonic acid with small amounts of neutral sugars present.

The degradation pattern of F4-I differed from DEAE F4-S. Only small amounts of material eluted in the polysaccharide region with the majority eluting as small molecular fragments (K_{av}= 0.69-1.0). Neutral sugar composition of these fractions were essentially the same as those found in the F4-S degradation products.

The HW extract of alfalfa stems is a complex mixture of carbohydrates. The majority of polysaccharides would be classified as rhamnogalacturonans (polygalacturonic acid backbone with intramolecular substitutions of rhamnose). This general group can be divided into two subgroups (F4-S & F4-I). Both polysaccharide groups contain methyl esters that block degradation by the endo-polygalacturonase. In F4-S the rhamnose and some of the galacturonic acid residues are branched containing large amounts of neutral sugars (arabinose and galactose). These substitutions are polysaccharide in nature based on the HW-55 elution pattern. This extensive substitution also inhibits degradation. Removal of the neutral sugars by mild acid hydrolysis followed by enzyme treatment eliminates the neutral polysaccharide fraction (K_{av}=0.19-.43) with the total carbohydrate eluting as small fragments (K_{av}=0.82-0.97). The general structure of this molecule contains regions which have high amounts of rhamnose with arabinans and arabinogalactans attached followed by regions of uninterrupted galacturonic acid residues which are methyl esterified. In F4-I there is much less substitution on either rhamnose or galacturonic acid residues. The structure of this group would be a rhamnogalacturonan backbone with minor neutral sugar substitution. Nearly all the galacturonic acid residues must contain methyl esters.

[1]$K_{av}=(V_e-V_o)/(V_t-V_o)$ V_e=sample elution volume, V_o=void volume, V_t=total included volume.

CORE AND NON-CORE LIGNIN COMPONENTS OF DIVERGENT SMOOTH BROMEGRASS GENOTYPES

H.G. JUNG and M.D. CASLER

Introduction

Lignin is the component of plant cell walls that is generally regarded as the primary factor limiting extent of forage fiber digestion by ruminants. In recent years it has been recognized that composition of lignin is variable and that composition features are correlated with fiber fermentability. Lignins have been characterized as core lignin: the high molecular weight, highly condensed polymer; and non-core lignin: esterified phenolic monomers. Concentration of core lignin and proportional contents of nitrobenzene oxidation products of core lignin have been shown to be negatively

correlated to in vitro fiber fermentability. Esterified non-core lignin components have been found to be negatively, p-coumaric acid, and positively, ferulic acid, correlated with fiber digestibility. The relative importance of these different lignin fractions as limitors of ruminal cell wall degradability is unknown. The objective of this study was to characterize the lignins of smooth bromegrass genotypes selected for divergent in vitro dry matter disappearance (IVDMD).

Materials and Methods

Nine smooth bromegrass genotypes selected for high IVDMD and nine selected for low IVDMD were grown in a randomized complete block nursery with 4 field replicates. Plants were harvested at the heads-emerged maturity stage in 1984 and 1985. Samples were separated into leaf blade and stem plus sheath fractions. Core lignin concentration was estimated by the sequential detergent procedure using 72% sulfuric acid. Forage samples were pre-extracted with neutral detergent prior to sequential extraction of esterified non-core lignin with anaerobic 1N NaOH and core lignin components by alkaline-nitrobenzene oxidation. Phenolic monomers released by these treatments were identified and quantified by reverse-phase LC.

Results and Discussion

The high IVDMD smooth bromegrass genotypes had lower concentrations of core lignin than low IVDMD genotypes in stem tissue, but not leaves (Table 1). Yield of core lignin components by nitrobenzene oxidation was greater from the leaf tissue of the low IVDMD group than the high group. Proportions of p-coumaric acid and syringaldehyde in the core lignin components were greater for the high IVDMD genotype. Total esterified phenolic yield from the high IVDMD group leaves was greater than for the low group, with ferulic acid content being responsible for the difference (Table 2). Although total esterified phenolics of stems did not differ, ferulic acid concentration of the high group was greater than the low IVDMD genotypes. The large difference in total esterified phenolics between leaf and stem fractions was primarily due to six-fold higher levels of p-coumaric to ferulic acid in the direction of higher relative concentrations of p-coumaric acid in less digestible tissues; stems vs. leaves, low vs. high IVDMD genotypes; agrees with correlations noted previously, but the ratio may be rather meaningless because p-coumaric acid is esterified to core lignin and ferulic acid esterified to hemicellulose. It is not clear why a shift in the ratio of these non-core lignin components would cause a change in fiber degradability. Higher levels of esterified ferulic acid in more degradable tissue may reflect a shift away from ester-ether ferulic acid bridges between core lignin and hemicellulose.

The data clearly illustrate that smooth bromegrass that has been divergently selected for IVDMD differs both in core and non-core lignin concentrations and also in lignin fraction composition. The task remains to explain how these shifts in lignification can cause altered fiber digestibility. Successful description of these mechanisms may facilitate genetic improvement in forage quality by selection based on components of lignification.

Table 1. Lignin composition of IVDMD genotype selection groups[a].

IVDMD	Lignin	NBO	Molar proportion				
Group	g/kg NDF	g/kg lignin	PCA	FA	PHBA	VAN	SYAL
Leaf							
High	64	22*	.019*	.118	.033	.551	.279*
Low	64	27	.017	.101	.028	.589	.265
Stem							
High	48*	41	.062	.150	.040	.410	.339
Low	57	40	.055	.155	.040	.418	.332

[a]Abbreviations: nitrobenzene oxidation products (NBO); p-coumaric (PCA, ferulic (FA) and p-hydroxybenzoic (PHBA) acids; vanillin (VAN); syringaldehyde (SYAL).
*Significant difference between IVDMD selection groups ($P<0.05$).

Table 2. Esterified phenolic composition of IVDMD genotype selection groups.

IVDMD	Total	Esterified Phenolics (mg/kg NDF)					Ratio
Group	g/kg NDF	PCA	FA	PHBA	VAN	SYAL	PCA/FA
Leaf							
High	3.57*	736	2454*	27*	60	91	.36*
Low	3.22	733	2306	28	60	89	.38
Stem							
High	8.50	4458	3854*	20	85	86	1.38*
Low	8.29	4501	3596	20	87	81	1.51

*Significant difference between IVDMD selection groups ($P<0.05$).

CORRELATION OF LIGNIN FRACTIONS WITH FIBER DEGRADABILITY IN WARM-SEASON GRASSES

H.G. JUNG and K.P. VOGEL

Introduction

Lignin concentration in forages is generally found to be negatively correlated with fiber digestibility. The extent of which lignin explains variation in fiber digestibility declines as the forages in the sample set become more diverse. The inhibition of fiber digestion by lignin in grasses appears to be much greater than for legume lignin. Grasses and legumes differ for both concentration and composition of lignin fractions. The relative importance of core lignin concentrations vs. non-core lignin, and lignin composition as predictors of forage fiber digestibility has not been assessed. The objective of this study was to determine which components of lignification were correlated with fiber digestion in warm-season grasses harvested at several stages of physiological maturity.

Materials and Methods

Five cultivars of switchgrass and 4 cultivars of big bluestem were grown in a randomized complete block nursery with 3 field replicates. Forages were harvested at the vegetative, boot and heading stages of maturity. After separating into leaf tissue and stem plus sheath fractions, forage samples were analyzed for core lignin concentration by the sequential detergent system with 72% sulfuric acid. Anaerobic 1 M NaOH was used to extract esterified non-core lignin phenolics from 80% ethanol insoluble preparation of the forage materials. The alkali-insoluble residues were treated with alkaline-nitrobenzene to determine core lignin composition and etherified non-core lignin phenolics. Phenolics were identified and quantified by LC. Fiber digestibility was measured as NDF disappearance after a 48 h in vitro fermentation with ruminal microorganisms.

Results and Discussion

Big bluestem exhibited lower NDF digestibility than switchgrass of both leaves and stems at the vegetative stage of maturity, but big bluestem fiber was more digestible at later stages of maturity. Switchgrass leaf tissue contained less core lignin than big bluestem leaves, but stem tissue of switchgrass accumulated more core lignin than did that of big bluestem. Ester- and ether-linked non-core lignin concentrations were consistently greater in switchgrass than big bluestem. Core lignin of switchgrass contained a higher molar percentage of syringaldehyde than big bluestem.

While core lignin concentration was negatively correlated with NDF digestibility across maturity stages (Figure 1), within a maturity stage and forage species core lignin concentration was not correlated with fiber digestibility. Only for big bluestem stems, in the vegetative stage of development, was core lignin significantly ($P<0.10$) correlated with NDF digestibility. Even across maturity stages, as shown for stems in Table 1, core lignin concentration was not correlated with NDF digestion for switchgrass. Within the maturity stages, esterified ferulic acid was found to be both positively and negatively correlated with NDF digestibility, yield of nitrobenzene oxidation products of core lignin was positively correlated with NDF digestibility, and the molar proportion of syringaldehyde in core lignin was negatively related to fiber digestion. There was no obvious pattern in the data as predictive variables changed between forage species and among maturity stages.

It is evident that more information is required before measures of lignin composition can be effectively used to predict forage digestibility. However, the data also clearly show that core lignin concentration is, at best, only a predictor of forage maturity and is not a good predictor of fiber digestibility within maturity stage. Therefore, lignin concentration would probably be a poor selection criterion for a forage improvement program if plant maturity is held constant.

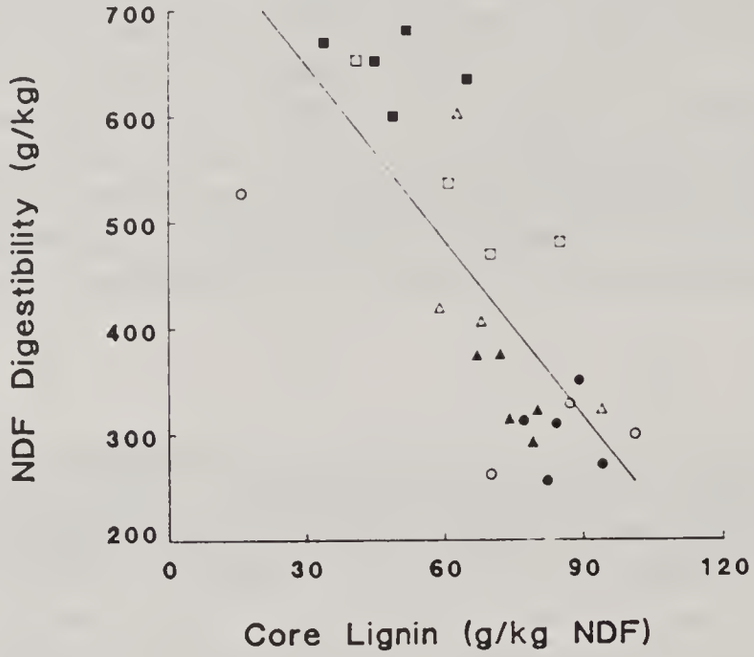


Figure 1. Relationship of in vitro NDF digestibility with core lignin concentration for switchgrass (closed symbols) and big bluestem (open symbols) when harvested at vegetative (❑,■), boot (Δ,▲) and heading (O,●) stages of maturity.

Table 1. Best single variable for predicting stem NDF digestibility of warm-season grasses. Sign in parentheses shows direction of variable's effect.

		Maturity		
Species	Overall	Vegetative	Boot	Heading
Switchgrass	FA (+)***	FA (-)***	NS	NS
Big bluestem	LIG (-)**	PSYAL (-)*	NBO (+)*	NBO (+)***
Overall	LIG (-)***	FA (+)***	FA (+)**	NBO (+)***

Abbreviations: Core lignin (LIG); esterified ferulic acid (FA); nitrobenzene product yield from core lignin (NBO); molar proportion syringaldehyde in core lignin (PSYAL).
*, ** and *** P<0.10, 0.05 and 0.01, respectively.
NS=non-significant (P>.10).

RELATIVE IMPACT OF DIGESTION RATE AND DIGESTION LAG ON TOTAL CELL-WALL DIGESTION

D.R. BUXTON

Introduction

Digestion of herbage by ruminants is limited primarily by concentration of neutral-detergent fiber (NDF) in herbage and degradability of this cell-wall material. On the basis of digestibility, NDF can be divided into two fractions: potentially digestible NDF (PDNDF) and indigestible NDF (IDNDF); which is not digested regardless of length of fermentation. Digestion of NDF can be characterized by a two-stage process involving a lag followed by microbial degradation. Both duration of lag and digestion rate of PDNDF determine the time required for PDNDF digestion. The digestion lag is probably related to hydration rate of NDF and/or time needed for microbial association with NDF. Digestion rate of PDNDF seems to be related to intrinsic chemical characteristics of the NDF. This study was undertaken to determine the influence of digestion lag and rate of digestion on total digestion time of PDNDF.

Materials and Methods

Immature and mature stems or reproductive tillers of alfalfa, birdsfoot trefoil, smooth bromegrass, and orchardgrass were collected in late May and mid July from four field replicates. At the early harvest, legumes were in the late-bud or early flower stage of maturity, and grasses were undergoing stem elongation with some flag leaves visible. At the late harvest, the plants were in the early- to midseed-ripening stages of maturity. The basal 150 mm of the main stem of harvested herbage was saved, and leaves, including sheaths, were removed. In vitro NDF digestibility of stems was determined after 0, 6, 12, 24, 36, 48, and 72 h of fermentation. These data were fitted with a first-order, nonlinear digestion model. Simple correlation coefficients were calculated among grass (n=48), legume (n=60) and both grass and legume (n=108).

Results and Discussion

The time required to digest 95% of PDNDF was similar for grasses and legumes when stems were immature, but more than doubled with maturity in grasses compared with an approximately 50% increase with maturity in legumes (Table 1). After 48 h of fermentation, 96 to 99% of PDNDF was digested in immature stems, whereas in mature stems an average of only 78 and 91% of PDNDF was digested (Table 1) in grasses and legumes, respectively.

Digestion rate of PDNDF averaged 0.085 h^{-1} when stems were immature. In mature stems, the rate decreased to 0.068 h^{-1} for legumes and 0.036 h^{-1} for grasses. Correlations coefficients between digestion rate of PDNDF and time for 95% of PDNDF to be digested were -0.83 for legumes, -0.87 for grasses, and -0.84 for both legumes and grasses. Similar coefficients between lag and time for 95% of PDNDF to be digested were -0.44, 0.16, and -0.13. Hence, digestion rate of PDNDF had a greater influence on variation in time required for 95% of PDNDF to be digested than did lag.

Table 1. Time required for 95% of potentially digestible NDF (PDNDF) to be digested and proportion of potentially digestible NDF that was digested after 48 h of fermentation.

	95% digestion			PDNDF digested in 48 h		
	Year 1	Year 2		Year 1	Year 2	
Species/cultivar	Mature	Immature	Mature	Mature	Immature	Mature
	h			%		
Smooth bromegrass						
Barton	79.2	39.9	101.1	82.6	96.7	72.2
Rebound	122.5	45.6	101.3	68.8	94.9	72.4
Mean	100.8	42.8	101.2	75.7	95.8	72.3
Orchardgrass						
Napier	83.9	33.4	85.0	81.5	98.2	79.1
Orion	77.5	42.7	75.2	83.9	95.7	83.0
Mean	80.7	38.0	80.6	82.7	97.0	81.0
Grass mean	90.8	40.4	90.9	79.2	96.4	76.6
Alfalfa						
Magnum	43.3	40.3	90.9	95.2	96.3	88.0
Spredor 2	51.2	33.0	65.4	93.2	98.3	86.5
Mean	47.2	36.6	78.2	94.2	97.3	87.2
Birdsfoot trefoil						
Empire	29.6	38.5	53.2	98.5	96.9	91.3
Viking	33.6	49.7	48.2	96.9	92.4	93.3
Mean	31.6	44.1	50.7	97.7	94.7	92.3
Red Clover						
Arlington	92.5	28.0	57.8	79.6	98.8	90.4
Legume mean	50.0	37.9	57.6	92.7	96.5	89.9

DIGESTIBILITY BY RUMEN MICROORGANISMS OF CELL WALL CARBOHYDRATES IN FORAGE STEMS AND LEAVES

D.R. BUXTON

Introduction

Neutral and pectic sugars, as well as isolated polysaccharides, are nearly completely digestible by rumen microorganisms. However, degradation of similar polysaccharides in the cell wall matrix of plants is rarely complete. The rate and extent to which they are digested varies with plant species, tissue, and maturity. Major factors limiting digestibility of polysaccharides in

walls of plants is bonding and shielding by lignin, and hydrogen bonding among polysaccharides. These limitations are more apparent in stems than in leaves, and in mature tissues than in immature tissues. Only limited information exists regarding which polysaccharide fractions are most affected when potential digestibility varies in forages.

The manner in which cell wall polysaccharide digestibility is affected by maturity probably varies between grasses and legumes because of the manner in which lignification proceeds. In stem bases of grasses, lignification of cell walls doubles as plants mature after the stem elongation stage. During this same period, lignification of legume cell walls increases by only 15 to 20%. The objective of this investigation was to determine the relationships among carbohydrate digestibility within grasses and legumes.

Materials and Methods

Immature and mature stems, and leaves of immature plants, of 'Magnum' alfalfa, 'Viking' birdsfoot trefoil, 'Rebound' smooth bromegrass, and 'Napier' orchardgrass were collected on common dates during 2 years in late May and again in late June or early July from four field replicates. Stem sections were of the 200 mm of basal stem after harvest near the soil line. Ground samples were incubated in buffered rumen fluid for 0, 6, 12, 24, 48, and 72 h. Neutral detergent fiber, acid detergent fiber (ADF), and sulfuric acid lignin were determined by using an amylase modification of the sequential procedure. Neutral sugars in NDF residues were determined by gas-liquid chromatography. To determine digestion rates, concentrations of NDF, cellulose (ADF minus lignin-plus-ash), hemicellulose (NDF minus ADF) and neutral sugars in NDF were fitted with a first-order, nonlinear digestion model or a linear model after logarithmic transformation.

Results and Discussion

In stems, lignin concentration in NDF increased from 176 to 209 g kg^{-1} (19%) with maturity in legumes compared to an increase from 53 to 102 g kg^{-1} (92%) in grasses. The digestion model revealed that potentially digestible NDF in stems decreased from 371 to 289 g kg^{-1} NDF (22%) with maturity in legumes and from 476 to 254 g kg^{-1} (47%) in grasses. The larger decrease in grasses was caused by a 53% decrease in potentially digestible cellulose compared to a 22% decrease in potentially digestible cellulose in legumes.

Digestibility of neutral sugars generally was greatest in leaves, followed in turn by immature stems and mature stems (Table 1). With 72 h of fermentation, 77 to 89% of the glucose was digested in leaves compared to 51 to 70% in immature stems and 29 to 41% in mature stems. Glucose digestibility decreased by 30% with stem maturity in legumes and by 49% in grasses. Xylose had the lowest digestibility of the neutral sugars in both stems and leaves. Its digestibility was particularly low in legume stems where less than one-fourth of the xylose was digestible. By comparison, approximately three-fourths of the xylose was digestible in legume leaves, nearly two-thirds in grass leaves, half in immature stems, and less than one-fourth in mature grass stems. This marked change in xylose digestibility was nearly paralleled by change in glucose digestibility. Approximately 95% of arabinose was digestible in legume leaves and slightly less in grass leaves. Nearly 80% was digestible in legume stems and somewhat less in grass stems.

Galactose digestibility was near 90% in both grass and legume leaves. It was the only neutral sugar to show a greater digestibility decline in legume than in grass stems. Mannose was completely digestible in grass leaves and nearly so in legume leaves and in grass stems. Rhamnose digestibility was as high in stems as in leaves.

Differences in neutral sugar digestibilities gives insight into the nature of lignin and phenolic bonding, and shielding of cell wall carbohydrates. Lignin, p, coumaric acid, and ferulic acid are thought to be linked through arabinose side chains of arabinoxylan, at least in grasses. The high digestibility of arabinose in this study suggests that only a small fraction of this neutral sugar is bonded to phenolic or lignin, even in highly lignified mature stems. In contrast, the low digestibility of xylose, particularly in legume stems, suggests that it plays a more important role in bonding to lignin or phenolics and protecting of cellulose. Whereas arabinose comprised only 2 to 4% of the neutral sugars, xylose comprised 18 to 30% of the neutral sugars in stems and from 11 to 27% in leaves. Thus, the role of xylose in limiting cell wall carbohydrate digestibility may be much greater than that of arabinose.

Table 1. Digestibility of neutral sugars from cell walls of leaves, immature stems, and mature stems following 72 h of fermentation in rumen fluid.

Species/part	Glucose	Xylose	Arabinose	Galactose	Mannose	Rhamnose
	.....	.....	g kg^{-1}	.....	.....	.....
Legumes						
Leaves	841	759	955	941	807	782
Immature stems	556	235	804	734	736	788
Mature stems	388	179	812	481	702	811
Grasses						
Leaves	804	625	872	917	1000	—
Immature stems	618	553	705	959	853	—
Mature stems	316	245	642	859	908	—
Significance						
Leaves						
Leg. vs. grs.	NS	NS	*	NS	NS	—
Stems						
Leg. vs. grs.	NS	*	NS	**	*	—
Maturity	**	**	NS	**	NS	NS
Leg. vs. grs.	NS	NS	NS	NS	NS	—

NS=not significant; *, **=significant at 0.05 and 0.01 levels, respectively.

NEUTRAL DETERGENT FIBER DIGESTION RATES OF ALFALFA RELATED TO GROWTH TEMPERATURE

R.A. LEWANDOWSKI and R.P. WALGENBACH

Introduction

Within a season in the Midwest, alfalfa grows under a wide range of temperatures. Raising the growth temperature of alfalfa accelerates a change from vegetative to reproductive growth and development. Alfalfa growth temperature also can affect the chemical constituent concentration and the extent of in vitro dry matter disappearance of alfalfa herbage, as well as anatomical and morphological characteristics. These growth temperature effects are mediated via effects on physiological processes that may alter metabolism of specific compounds and/or patterns in combining compounds that ultimately alter anatomy, morphology and chemical make-up. Such temperature induced changes also may affect the way in which rumen microbes degrade alfalfa herbage. Our objectives were to investigate if alfalfa growth temperatures affected the rate of neutral detergent fiber digestion of alfalfa leaves and stems.

Materials and Methods

Four replicates of alfalfa lines selected for rapid, moderate and slow regrowth rates were grown in separate chambers for two growth periods at day/night temperature regimes of 16.5/8, 24.5/16.5 and 32.5/24°C. The desired harvest (H) maturity stages for each of the two growth periods were early to late bud (ELB), late bud to first flower (LBFF), and early to late flower (ELF). Regrowth lines were combined within maturity stages. Finely ground leaves and stems were digested in vitro with rumen liquor for 0, 5, 10, 15, 20 and 72 h. Following incubation, fiber was extracted sequentially by neutral and acid detergents. These data were fitted with the first order, nonlinear digestion model.

Results and Discussion

The H1 stems of ELB alfalfa had a faster rate of NDF digestion (0.110 h^{-1}) than did stems of LBFF (0.097 h^{-1}) and ELF (0.090 h^{-1}) alfalfa. The H2 stems had NDF digestion rate trends similar to those of H1. The NDF digestion rates differed little among maturity stages for H1 and H2 leaves. Growth temperatures did not affect NDF digestion rates for finely ground alfalfa stems (Table 1). The leaves from alfalfa grown at 16.5/8°C had a faster rate of NDF digestion in H1 and H2 than did those grown at 24.5/16.5 and 32.5/24.5°C. Growth temperature induced effects on NDF digestion rates of leaves could be useful in studying overall fiber digestion kinetics. Fine grinding of stems may have prevented detection of potential differences in NDF digestion rates of alfalfa grown at the different temperatures.

Table 1. Digestion lag and rate of potentially digestible neutral detergent fiber in leaves and stems of alfalfa grown at day/night temperatures of 16.5/8, 24.5/16.5 and 32.5/24.5°C.[+]

	Leaves		Stems	
Temperature,°C	Lag	Rate	Lag	Rate
	h	h^{-1}	h	h^{-1}
		HARVEST 1		
16.5/8	1.1	0.229	1.8	0.108
24.5/16.5	1.2	0.073	3.3	0.990
32.5/24.5	2.1	0.056	2.3	0.090
LSD 0.05	NS	0.078	1.0	NS
		HARVEST 2		
16.5/8	1.3	0.123	2.8	0.086
24.5/16.5	2.6	0.092	3.2	0.098
32.5/24.5	3.3	0.066	4.3	0.129
LSD 0.05	0.7	0.029	NS	NS

[+]Means of three regrowth types and 3 maturity stages.

CROP-WATER-STRESS INDEX AND FORAGE QUALITY RELATIONSHIPS IN ALFALFA

R.A. HALIM, D.R. BUXTON, M.J. HATTENDORF and R.E. CARLSON

Introduction

Water deficits cause stomatal closure, reduced transpiration, and elevated canopy temperature. Measurement of canopy temperature to asses plant-water stress has become increasingly common. Hand-held infrared thermometers provide precise and quick measurements and allow numerous non-destructive readings to be taken. The Crop Water Stress Index (CWSI), based on canopy-minus-air temperature (Tc-Ta), has been used to assess relationships of alfalfa forage yield under water-deficit stressed conditions. The increase in Tc-Ta from decreased plant-water potential is linear to -3.0 MPa. A CWSI of 0 indicates that the crop is not stressed and transpiring at its potential rate. A CWSI of 1.0 indicates that water stress is maximal and that transpiration has ceased. Although the CWSI has been developed to predict forage yields, it also may be related to forage quality. Numerous studies have shown that nutritive quality of alfalfa changes with intensity of water-deficit stress. Our objective was to assess the relationship between the CWSI and forage quality traits for plant parts and total herbage of alfalfa grown under water deficit.

Materials and Methods

'Apollo II' alfalfa was grown in 100-L photometers set into the ground and protected by a movable rain-out shelter. Plants were watered either weekly or twice weekly at 112, 100, 88, 76, and 64% of field capacity during 2 years. Daily readings of canopy and air temperature were taken and vapor-pressure deficits were determined from the time plants were 15-cm tall until harvest. Plants were harvested after 5 weeks regrowth and were divided into stem bases (below 6 nodes), stem tops, and leaves before forage quality analyses were conducted.

Results and Discussion

Dry matter yield and plant maturity declined linearly with increasing CWSI (r^2 = 0.86 and 0.65, respectively). Leaf-to-stem ratio increased with increasing CWSI (r^2 = 0.51), but year effects, in addition to water stress effects, were evident. In-vitro digestible dry matter (IVDDM) in stem bases increased with CWSI (Figure 1) but leaf IVDDM was not significantly related to CWSI. Crude protein (CP) concentration is stem bases increased with CWSI (Figure 2), whereas leaf CP concentration declined in a quadratic manner with increasing CWSI (Figure 3). Total-herbage concentration was not significantly related to CWSI. Neutral-detergent fiber concentration in stem bases, leaves, and total herbage declined with increasing CWSI (r^2 = 0.89, 0.75, and 0.69, respectively.)

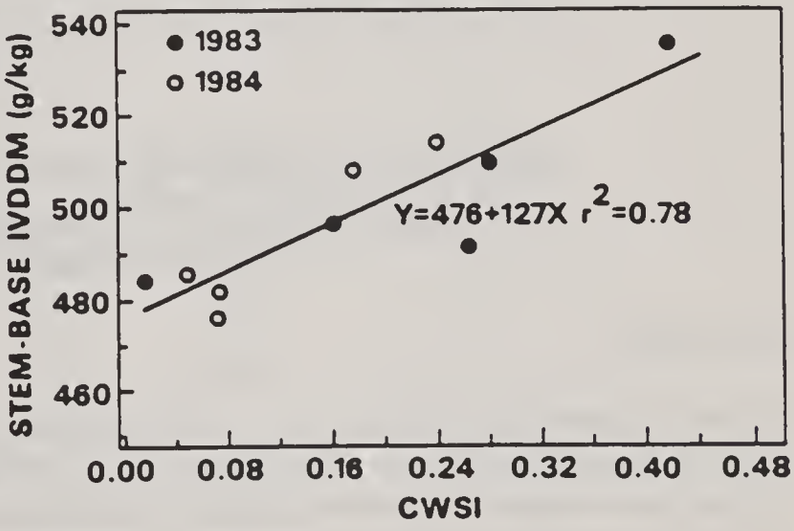


Figure 1. Regression of in vitro digestible dry matter (IVDDM) in stem bases from plants grown in 2 yr on Crop Water-Stress Index (CWSI).

That CWSI was more closely related to dry-matter yield than to most forage quality traits underscores that CWSI was designed primarily for predicting yield responses rather than quality to water stress. Total-herbage quality is influenced by the quality of both leaves and stems, and because these components do not always respond in the same manner to water stress, total-herbage quality cannot easily be related to the CWSI. Neutral-detergent fiber concentration was the only parameter that was consistently related to CWSI for all plant parts and total herbage.

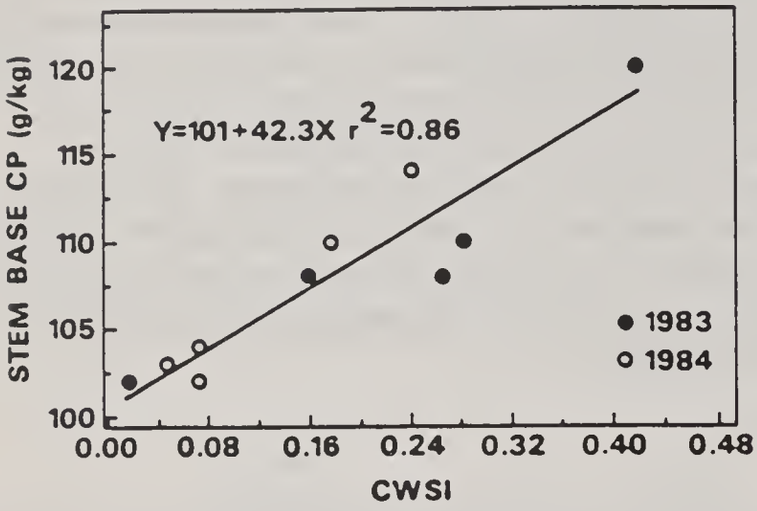


Figure 2. Regression of crude-protein (CP) concentration in stem bases from plants grown in 2 yr on Crop Water-Stress Index (CWSI).

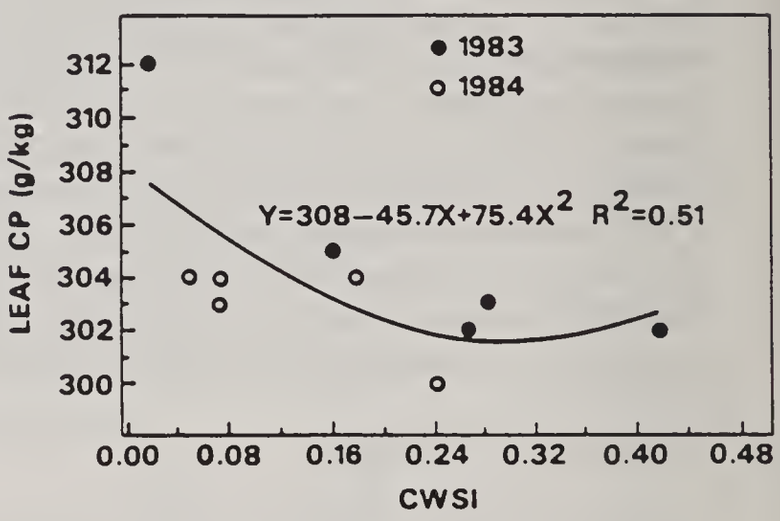


Figure 3. Regression of crude-protein (CP) concentration in leaves from plants grown in 2 yr on Crop Crop Water-Stress Index (CWSI).

SELECTION FOR HIGH AND LOW DIGESTIBILITY IN LIGNIN SELECTED ALFALFA POPULATIONS

C.S. ENDRES and R.R. SMITH

Introduction

One of the greatest limitations to the nutritive value of alfalfa is the digestibility of the stem tissue of the plant, especially the basal portion of the stems. Previous research has shown that the digestibility of alfalfa stem tissue decreases as the age of the tissue advances. However, some evidence exists that would suggest that genetic variability for digestibility of alfalfa stem tissue may be present at any specific stage of plant development. The objective of this study was to apply divergent selection on stem in vitro digestible dry matter (IVDDM) in high and low lignin populations of alfalfa.

Materials and Methods

Using NIRS technology, IVDDM values were estimated on basal stem tissue of two-year-old plants in high and low lignin (HL and LL) alfalfa populations. Within each population both high digestible (HD) and low digestible (LD) plants were identified and intercrossed to produce four populations: high lignin - high digestibility (HLHD); high lignin - low digestibility (HLLD); low lignin - high digestibility (LLHD); and low lignin - low digestibility (LLLD). Progeny plants representing each derived population were transplanted to a replicated test in the field, sampled twice (Cut I and Cut II) at the bud stage of growth, and analyzed (basal stem tissue) for IVDDM.

Results and Discussion

Mean digestibility of the progeny in each derived population regressed back toward the overall mean of the two original lignin populations (data not presented). However, selection for digestibility was effective in the high lignin population (Table 1.). The difference between the mean of the high and low digestible derived populations in the high lignin population was significant at both harvests. Thus selection was effective in separating the high lignin population into a high and a low digestible group. On the other hand, significant variation among progeny families was only detected in the material selected for low digestibility in both lignin populations. Parent-offspring correlations (a measure of heritability) for IVDDM was significant in the second harvest ($h^2 = 0.66$), but not considered to be very high. While divergent selection was effective in one population, continued selection for IVDDM may be limited in these highly specialized alfalfa populations. One might expect to be more successful by applying similar selection in more adapted alfalfa germplasm.

Table 1. Response to selection for IVDDM in high and low lignin alfalfa populations in 1988 at Arlington, WI.

Harvest	Popn.	HD	LD	Diff. (HD-LD)
		.....g kg^{-1}.....		
Cut I	HL	450	435^{++}	15*
	LL	454	449^{++}	5
Cut II	HL	431	408	23**
	LL	433	426	7

$^{++}$Significant differences among families within a population.

*,**Difference is significant at the 5 and 1% level, respectively.

ESTIMATES OF QUALITY PARAMETERS IN RED CLOVER CULTIVARS AND SELECTED GERMPLASM

R.R. SMITH and D.K. SHARPEE

Introduction

Red clover is an excellent forage legume being adapted to a wide range of soil and management conditions. It is especially adapted to more acid, wet soils and to short rotation management systems and is excellent for pasture renovation. Estimates of parameters associated with forage quality are lacking in the species. Therefore, the following study was initiated to obtain estimates of neutral and acid detergent fiber (NDF and ADF) concentration and their interaction with season and year of forage production on red clover germplasm.

Materials and Methods

Sixteen midwest-adapted red clover cultivars and experimental strains were established in replicated trials on the Agronomy Experimental Farm, Arlington, WI in both 1984 and 1985. Herbage samples were taken at the bud stage of growth development at both first harvest (15 June) and at second harvest (25 July) in 1985 and 1986 on the 1984 established trial and in 1986 on the 1985 established trial. Samples were dried with hot forced-air at 50 C and ground through a 1mm screen in a cyclone mill. Ground herbage samples were analyzed for NDF and ADF.

Results and Discussion

The interaction of cultivars/strains with date of harvest, year of production, or trial was not significant for either quality parameter (NDF or ADF). Concentration of both NDF and ADF was significantly higher in the first harvest than the second harvest (396 vs 373 g kg^{-1} for NDF and 269 vs 227 g kg^{-1} for ADF) (Table 1). No significant variation was observed among cultivars/strains for either parameter for any one harvest indicating very little variation among cultivars/strains for these parameters. The range in performance among cultivars/strains ranged from 11 to 12% of the mean for NDF and from 13 to 19% of the mean for ADF. Average coefficients of variation were well within acceptable limits.

The lack of variability among red clover cultivars for NDF and ADF would suggest that the current germplasm is genetically quite similar for these quality parameters. Since the populations are genetically heterogenous, it still seems feasible to improve specific population for these parameters through appropriate selection.

Table 1. Mean NDF and ADF concentration (g kg^{-1}) across and among red clover cultivars/strains (means across two trials).

	NDF		ADF	
	Cut I	Cut II	Cut I	Cut II
Across cultivars/strains				
Mean	396	373*	269	227*
CV (%)	4.3	5.9	4.1	7.2
Among cultivars/strains				
LSD (5%)	ns	ns	ns	ns
Range (from -	376	354	255	206
to)	418	397	291	250
Units in Range	4.2	4.3	3.6	4.4
Range as % of Mean	11	12	13	19

* Difference between cuts significant at the 5% level.

ELECTROPHORETIC EXAMINATION OF ALFALFA SILAGE PROTEINS

D.E. GRUM, W.L. SHOCKEY and W.P. WEISS

Introduction

Forage proteins undergo rapid proteolysis after ensiling which causes increases in ammonia nitrogen and non-protein nitrogen concentrations. Degradation of plant proteins occurs within the first few days after harvest through action of plant and microbial proteinases.

Increased accuracy in evaluation of the nitrogenous fraction of ensiled crops will enable rations to be more accurately balanced to meet the nutritional requirements of animals. Further, knowledge of the type of protein degraded in the silo will direct the development and implementation of silage

additives for increased protein preservation efficiency. The objectives were to 1) characterize buffer soluble proteins from fresh alfalfa herbage and silage into molecular weight ranges using polyacrylamide gel electrophoresis (PAGE), and 2) determine effects of additives and length of ensiling time on proteins of alfalfa silage.

Materials and Methods

First cutting alfalfa was cut at late bud stage of growth, wilted for 24 hr (ca. 40% DM), then chopped (2-3 cm) with a forage harvester. Chopped forage (100 kg) was loaded into a mixer, treatment applied, and mixed for 10 min. Treatments were control (C); glucose (G, 50 g glucose/kg DM); ammonium hydroxide (AH, 25 g ammonium hydroxide/kg DM); and a formic acid-formal dehyde mixture (FF, 18.3 g [2:1 (90% HCOOH:37% HCOH)]/kg DM). The forage was ensiled into 10 x 60 cm PVC experimental silos. Samples were taken on day 0 and, on days 1, 2, 4, 7, 21, and 50 of fermentation. Analyses were dry matter, pH, total nitrogen, non-protein nitrogen, ammonia nitrogen, organic acids (lactic, acetic, and butyric) water soluble sugar, NDF, ADF, and PAGE analysis of buffer soluble protein.

Results and Discussion

Fermentation as estimated by pH, organic acids, soluble sugar, and nitrogen fractionation data proceeded as expected. Ammonium hydroxide treatment had the highest initial pH (8.0), exhibited the most rapid decline in pH, and had the highest pH by day 50 of fermentation when compared to the mean of other treatments (4.87 vs 4.42).

Ammonia and non-protein nitrogen in AH treated forage increased at a lower rate compared to other treatments, and when expressed as percent of total nitrogen was higher than other treatments because of the ammonia contribution by the treatment. Concentrations of ammonia nitrogen for F/F treatment were lower than for C and AH, indicating reduced metabolism of protein breakdown products.

Proteins from alfalfa herbage and silage were divided into three groups based on relative mobility: $\geq$ 40 kDa (.019-.239 R_f), 40.6 - 12.3 kDa (.239-.564 R_f), and $\leq$ 12.3 kDa (.564-.800 R_f). Assuming all nonprotein nitrogen was extracted with buffer, and that 20% of the total forage nitrogen was nonprotein nitrogen, 36-40% of the total plant protein nitrogen was extracted.

Conclusions

Electrophoretograms demonstrated that proteolysis was inhibited by F/F and AH treatments but not G. Formic acid/formaldehyde decreased amount of extractable proteins by decreasing their solubility in the extraction buffer. Further research to extract a higher proportion of total plant protein and to determine significance of individual proteins to ruminant nutrition is warranted.

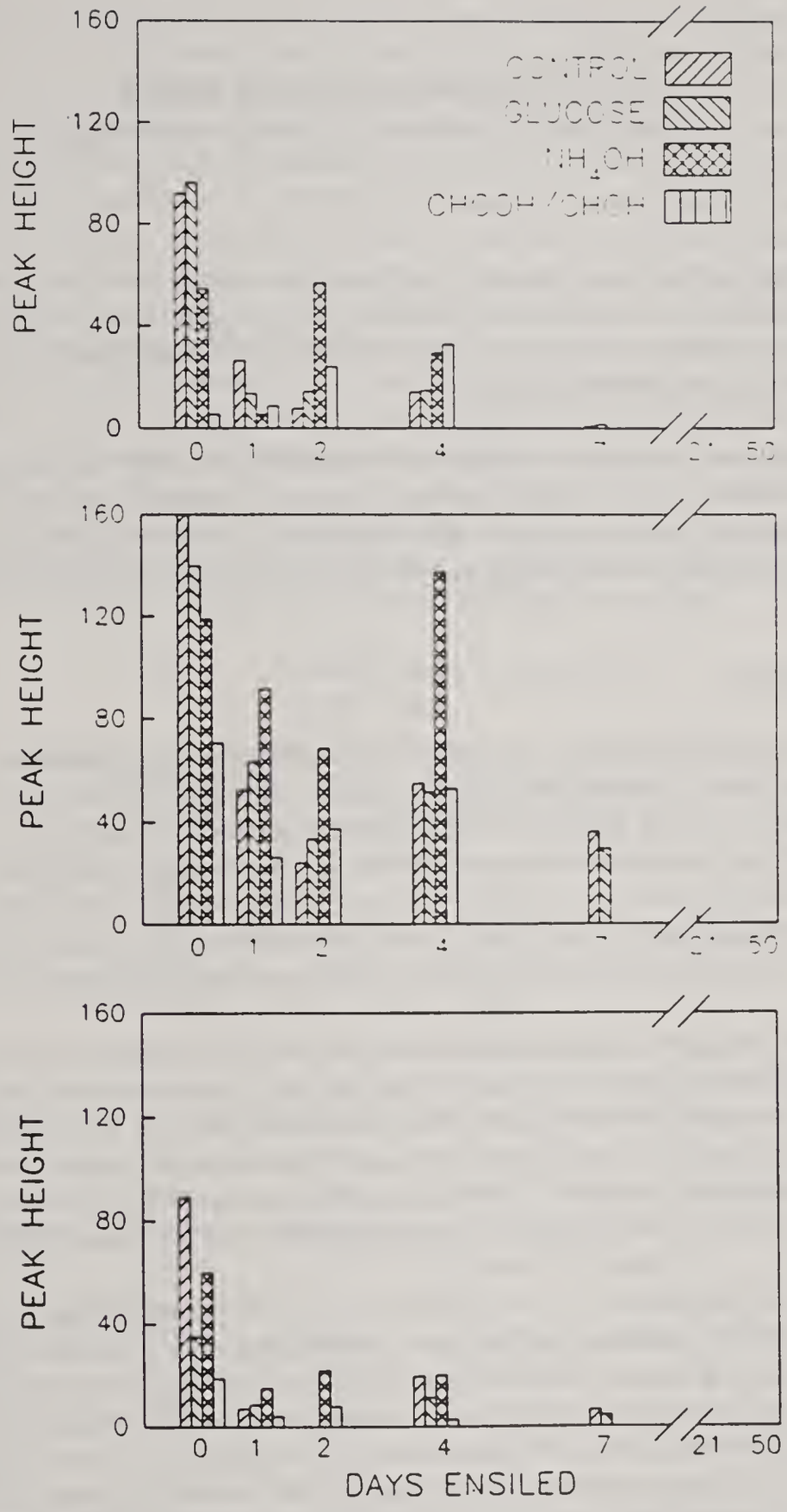


Figure 1. Relationship between the peak height of large (top), medium (middle), and small (bottom) molecular weight proteins and time for each treatment.

QUICK-DRYING FORAGE MATS

R.G. KOEGEL, R.J. STRAUB and K.J. SHINNERS

Introduction

Mats made from alfalfa at the time of mowing and placed on stubble have been shown in earlier research to dry to a moisture content suitable for baling in 2 1/2 to 6 hours under favorable conditions. Furthermore, alfalfa so harvested proved to have more rapid and extensive NDF digestion than conventionally harvested material.

Efforts during the past year have concentrated on: (1) harvest technology (2) preparation of mat-harvested and conventional alfalfa silage for a feeding trial of lactating cows which was conducted Jan-Mar. 1990 and (3) evaluation of an alternative maceration process based on simpler equipment which is also less vulnerable to foreign objects. Results of the last two studies will be covered in subsequent reports.

Material and Methods

In order to pick up the mat dried alfalfa from the stubble with minimum disturbance, a belt pickup unit was designed and built. This unit was similar to those used to pick up windrowed grain for combining. It consisted of a 0.8 m (31.5 in) wide rubber belt to which plastic pick-up fingers were attached. This width was chosen to accommodate mats produced from a prototype forage mat machine which generally produces mats of 0.4 - 0.5 m (16-20 in) width. The average length (roll center to center) of the unit was 1.3 m (52 in). It was operated at a maximum inclination of 32 degrees. The pick-up unit was built with a support and drive wheel at the front of each side.

The pick-up unit was mounted on a modified double chop flail type forage harvester for silage making. This unit had its flail rotor removed and the belt pick-up unit was mounted to deliver the mat-dried alfalfa to the top of the cross auger which fed the blower unit. The pick-up unit was also mounted on a small rectangular baler. The conventional tine and drum pick-up was removed and the belt-type unit was mounted in its place. The bale chamber was modified to increase resistance to bale movement in order to obtain bales of the desired rigidity from the compliant mat material.

Bales were weighed and measured as well as cored for moisture determination. Handling losses were measured for bales by gathering and weighing material lost during handling. Pick-up losses were determined by picking up mat dried alfalfa that had been laid on a woven plastic tarp at the time of mat formation. Loss and harvested fractions were recovered for weight and moisture determination. Each tarp had a length of approximately 30 m (100 ft). Pick-up losses were determined for mats of moisture contents suitable for hay, and for silage. Bale chamber losses were determined using a suspended catch pan underneath the entire length of the bale chamber so that losses could be recovered and quantified. Side drapes were used to minimize wind loss.

Results and Discussion

The pick-up unit was also mounted on the modified double-chop forage harvester early in the summer of 1989 and was successfully used to harvest approximately 4 ha (10 acres) of mat-dried alfalfa for a feeding trial. The action of the cross auger and of the blower paddles and knives effectively broke up and chopped the material for blowing into a transport unit and for ensiling. No further modification of the pick-up unit or of the original modified forage harvester were required for successful operation. Use of this unit reduced complexity, cost and power requirement inherent in the conventional forage harvester. In general, the combination of belt pick-up and cross auger allows the harvesting of mats which are considerably wider than the original pick-up of the forage harvester or baler.

The belt-type pick-up unit performed well for the variety of conditions under which it was used. It was able to lift mat dried alfalfa successfully at both baling and ensiling moisture levels and did so with losses of 1.2 to 1.5 percent of the total dry matter (Table 1). In all trials, the number of plunger strokes per bale was considerably higher than encountered in usual baling due to the narrow mats formed by the prototype macerator and the light crop conditions. Plunger strokes ranged from 35 to 58 strokes for bales of approximately 0.8 m (30 in) length. The resulting bales were 20-50 percent more dense than those now made by many forage producers (Table 2). Mechanical losses do not appear to be higher than those now encountered in hay or silage production.

Table 1. Pickup losses for tarp-dried alfalfa mats.

Hay Moisture (% w.b.)	Pick-up Loss (% total dry matter)
53	1.2
47	1.5
20.7	1.4
15.8	1.5

Table 2. Bale and loss fractions for mat-dried alfalfa hay.

Hay Moisture (% w.b.)	Bale Density (lb/ft^3)	Bale as Stored	Bale Chamber Loss	Handling Loss
		(% total dry matter)		
24.5	12.4	96.0	3.4	0.5
23.5	11.8	95.5	4.0	0.6
12.0	10.0	92.3	7.0	0.6
15.3	11.1	94.8	4.5	0.7
17.8	10.7	93.3	5.5	1.2

EFFECT OF SWATH MANIPULATION ON LOSSES AND DRYING OF ALFALFA

R.J. DAVIS, C.A. ROTZ and D.R. BUCKMASTER

Introduction

Harvest of a high-quality alfalfa crop is heavily dependent upon weather conditions. Speeding the drying process can reduce the risk of heavy losses due to rain damage. In the northern U.S., the typical alfalfa hay making system includes mowing with a mower-conditioner, raking when the alfalfa reaches 30 to 40% moisture content and baling when the crop is dry enough to store. European influences in recent years have suggested adding a tedding operation to increase the drying rate of hay. Recently-marketed swath inverters (windrow turners) may also aid in hastening field drying. The objective of this research was to compare the drying rate, field curing time and losses of alfalfa hay made with a standard procedure (mower-conditioner followed by raking) to the same procedure with tedding or swath inversion.

Materials and Methods

Three field trials were conducted to evaluate the possible benefits of swath manipulation in alfalfa hay production under mid-Michigan weather conditions. Alfalfa was cut with a mower-conditioner which used a cutterbar for mowing and intermeshing rubber rolls for conditioning. All treatments were located adjacent to one another to reduce crop and field variation. In trials 1 and 2, alfalfa was laid as a wide (2.1 m) swath for field curing. The 5 treatments compared were (1) raking at 40% moisture on the expected day of baling, (2) tedding immediately after mowing and raking at 40% moisture, (3) tedding the morning after the day of mowing and raking at 40% moisture, (4) inverting at 50% moisture on the expected day of baling and raking just prior to baling, and (5) inverting at 30% moisture and raking just prior to baling. In trial 3, the tedder was not included and swath width varied from a wide swath (2.1 m) to a narrow swath (1.0 m). The 5 treatments compared were (1) narrow swath with no manipulation, (2) wide swath raked at 35% moisture, (3) wide swath inverted when the top of the swath reached 40% moisture and raked when the whole swath reached 30%, (4) narrow swath inverted when the top of the swath reached 40% moisture, and (5) wide swath inverted to a narrow swath (1.5 m) when the top of the swath reached 40% moisture. Actual moisture contents at the time of tedding, inverting or raking varied from the target moisture contents.

Tedding was done using a four-rotor tedder (Kuhn Model GF440) pulled at a speed of 6.4 km/h. The inverter (Pequea Model 786) was operated at 4.8 km/h. This machine picked up the swath and rotated it 90 to 180 degrees (depending on swath width desired) around a semi-circular platform. The hay dropped from the platform into a swath adjacent to the original swath, completely inverted. All raking was performed using a parallel-bar rake. After raking, a 0.5 m^2 frame was randomly placed in an area formerly covered by the swath. Loss was the remaining alfalfa particles collected inside the frame.

A drying constant was calculated for each swath of each treatment from the day length and the beginning and final moisture contents. Drying constants for treatments were compared using a random block experimental design where each day formed a block and each swath was a replicate. Linear contrasts were used to determine the significance of differences among treatments. Field curing times of each treatment were compared, with field curing time being the time between mowing and baling.

Results and Discussion

Mechanical treatments did not significantly affect alfalfa drying rate or field curing time (Table 1). Although significant differences in curing times were not found, the variation within trial 3 was noteworthy. The 2 to 5 hour variation among treatments could mean that some treatments would stay in the field overnight. If rain occurs, this would be very detrimental.

Loss with the use of swath inversion was not consistent among trials. In trials 1 and 2, the loss following inversion and raking was similar to that following raking alone (Table 1). In trial 3, the loss from the swath which was inverted and raked was significantly greater than the loss from the swath which was just raked. Therefore, under certain circumstances, the added operation of swath inversion may increase crop loss. When swath inversion was used in place of raking, losses were similar with either the rake or inverter. Tedding always increased crop loss. In first cutting (trial 1), tedding followed by raking gave a greater loss than inverting followed by raking. In second cutting (trial 2), tedding followed by raking gave a greater loss than inverting followed by raking or raking alone. Moisture content of the crop at the time of tedding or inverting did not affect the amount of loss.

Table 1. Field curing times, drying rates and losses of alfalfa when field cured in two swath widths with and without swath manipulation.

Treatment	Curing time (h)	Mean drying rate (h^{-1})	Dry matter loss (%)
Trial 1			
Raked only	26	0.159	1.3
Inverted, raked	26	0.188	0.7
Tedded, raked	25	0.164	1.8*
Trial 2			
Raked only	44	0.165	1.9
Inverted, raked	44	0.161	2.4
Tedded, raked	44	0.188	6.3‡
Trial 3			
Narrow swath, no manipulation	56	0.062	0.0†
Wide swath, raked	56	0.074	1.0
Wide swath, inverted, raked	51	0.114	2.5‡
Narrow swath, inverted	54	0.077	1.2
Wide swath, inverted to narrow	52	0.091	0.9

*Greater loss than inverting/raking by linear contrast ($p<0.01$).
†Less loss than other treatments of trial by linear contrast ($p<0.01$).
‡Greater loss than other treatments of trial by linear contrast ($p<0.01$).

COMPARISON OF DRYING RATE AND LEAF LOSS AMONG ROLL TYPE MOWER-CONDITIONERS

R.G. KOEGEL, K.J. SHINNERS and R.J. STRAUB

Introduction

More than one-half million mower-conditioners are currently in use in the U.S. Most of them are of the roll type. Both manufacturers and farmers are keenly interested in how the various roll types affect both drying rate and leaf loss.

Materials and Methods

Seven different mower-conditioners provided by five manufacturers and representing four different roll types or roll combinations (Table 1) were run side by side to compare the resulting drying rates and leaf loss.

For leaf loss determination, 100 feet of each resulting maximum attainable width swath was dropped on woven plastic tarp rolled out concurrently with mowing. Forage was carefully removed from the tarp at two 10 foot long locations with a five-tine fork (2 inch tine spacing). Material remaining on the tarp at these locations was defined as "leaf loss". Cross-sections of the remaining swath were picked up periodically over 48 hours with a Carter plot harvester to determine drying rates. Four such trials were run on July 20, 24, 26, and 31, 1989.

Results and Discussion

Average drying rate constants are shown in Table 2. The drying rate constants are not different at P=0.05. Nor do drying rate constants for rolls of the same type group together.

Average losses for the seven machines are shown in Figure 1. Again, these are not generally statistically different. The most important difference in losses was with harvesting date as shown in Figure 2. Average losses over all machines increased from 4.23% on 7/20 to 7.22% on 7/31 as the alfalfa matured from 50-75% bloom to 100% bloom. The small average differences observed between roll combinations of the same type may be due to differences in parameters such as roll speed, roll clearance, and specific roll force. The last parameter was set by a representative of the manufacturer in each case.

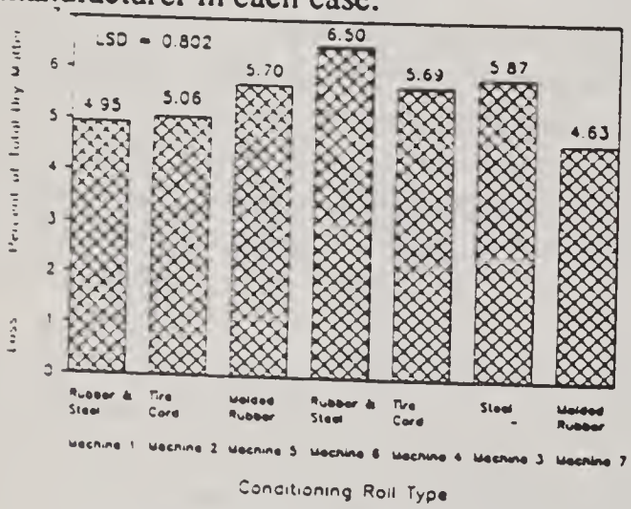


Figure 1. Average losses for the seven machines tested.

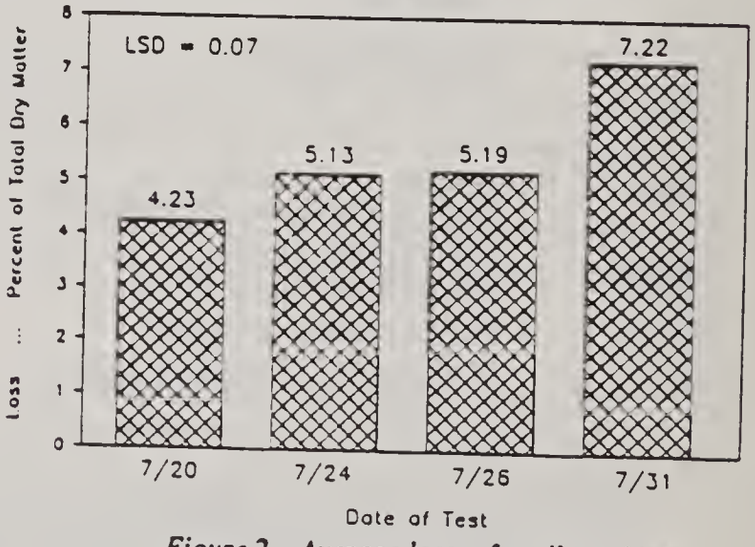


Figure 2. Average losses for all machines tested as a function of harvest date.

Table 1. Mower-conditioner machine specifications.

Machine No.	Upper Roll Type	Upper Roll Diameter cm	Lower Roll Type	Lower Roll Diameter cm	Intermeshing/ Fluted Rolls	Width of Cut cm	Roll Width cm
1[1]	Molded Rubber	20.3	Steel	19.7	IM	282	279
2[1]	Tire Cord	20.3	Tire Cord	20.3	IM	282	279
3[2]	Steel	24.1	Steel	24.1	IM	282	269
4[2]	Tire Cord	22.9	Tire Cord	22.9	IM	282	269
5	Molded Rubber	19.7	Molded Rubber	19.7	IM	282	277
6[3]	Molded Rubber	17.8	Steel	17.8	FL	279	173
7	Molded Rubber	26.4	Molded Rubber	26.4	IM	274	259

[1,2]Machines with the same superscript were the same model, but had different conditioning rolls.

[3]Machine had rotary disk type cutting mechanism. All other machines had conventional sickle type cutting mechanisms.

Table 2. Swath drying trial.

Machine No.	Roll Type	Drying Constant (h-1)	Moisture (percent wet basis) Initial	Final
1	Rubber and Steel	0.1706	75.3	27.8
2	Tire Cord	0.1513	75.9	25.8
3	Intermeshing Rubber	0.1654	73.4	27.7
4	Tire Cord	0.1630	74.8	24.6
5	Intermeshing Rubber	0.1556	75.5	23.1
6	Rubber and Steel	0.1495	75.0	26.3
7	Intermeshing Rubber	0.1705	75.0	24.2
Least Signf. Difference (LSD) (P=0.05)		0.0470	4.1	7.5

Averages within columns were not statistically significant at the 95% level.

ENERGY REDUCTION IN FORAGE HARVESTING: FRICTION

L.L. LEHMAN, R.G. KOEGEL and K.J. SHINNERS

Introduction

Reduction of energy requirements for forage harvesting can improve profitability by reducing fuel and capital costs and/or by improving the timeliness of harvesting. Since friction between forage and machine surfaces may account for 25-30% of the energy required by such machines as forage harvesters and blowers, reducing the coefficient of friction has the potential for decreasing energy requirements significantly.

Materials and Methods

A rotating table was used to determine the coefficient of friction between chopped alfalfa and six different materials, with and without limited water lubrication, for a range of velocities, normal forces, and moisture contents. Subsequently, a forage blower was modified by adding nozzles for water lubrication and by overlaying a replacement band with ultra high molecular weight (UHMW) polyethylene. The energy required during silo filling as a function of band material and level of water lubrication was determined.

Results and Discussion

The effects of normal pressure and velocity levels were found to be relatively minor compared to alfalfa moisture content, surface material, and absence or presence of water lubrication. Figure 1 shows the coefficient of friction between chopped alfalfa (54% moisture content w.b.) and six different surfaces with and without limited water lubrication. Generally, water lubrication reduced the coefficient to about one-third of the unlubricated level. For a machine in which 30% of the energy required is attributable to friction, this would result in reducing the energy requirement to about 80% of the unlubricated value. While the differences between surfaces were not as great, the coefficient of friction for UHMW polyethylene was less than 80% that of polished steel for either the lubricated or non-lubricated condition.

In the blower, water lubrication of 170 liters/hr reduced the specific energy requirement to 75% or less of the unlubricated value for both polished steel and UHMW polyethylene (Figure 2). Because of the modest amounts of water required to obtain a substantial energy reduction, it is felt that water lubrication of field-going machines with high friction losses, like the forage harvestor, may be desirable. In the blower the UHMW polyethylene did not result in reducing energy requirements relative to polished steel. It is speculated, based on the appearnace of the polyethylene surface, that its high rate of thermal expansion may have caused it to bulge and to interfere with the impeller.

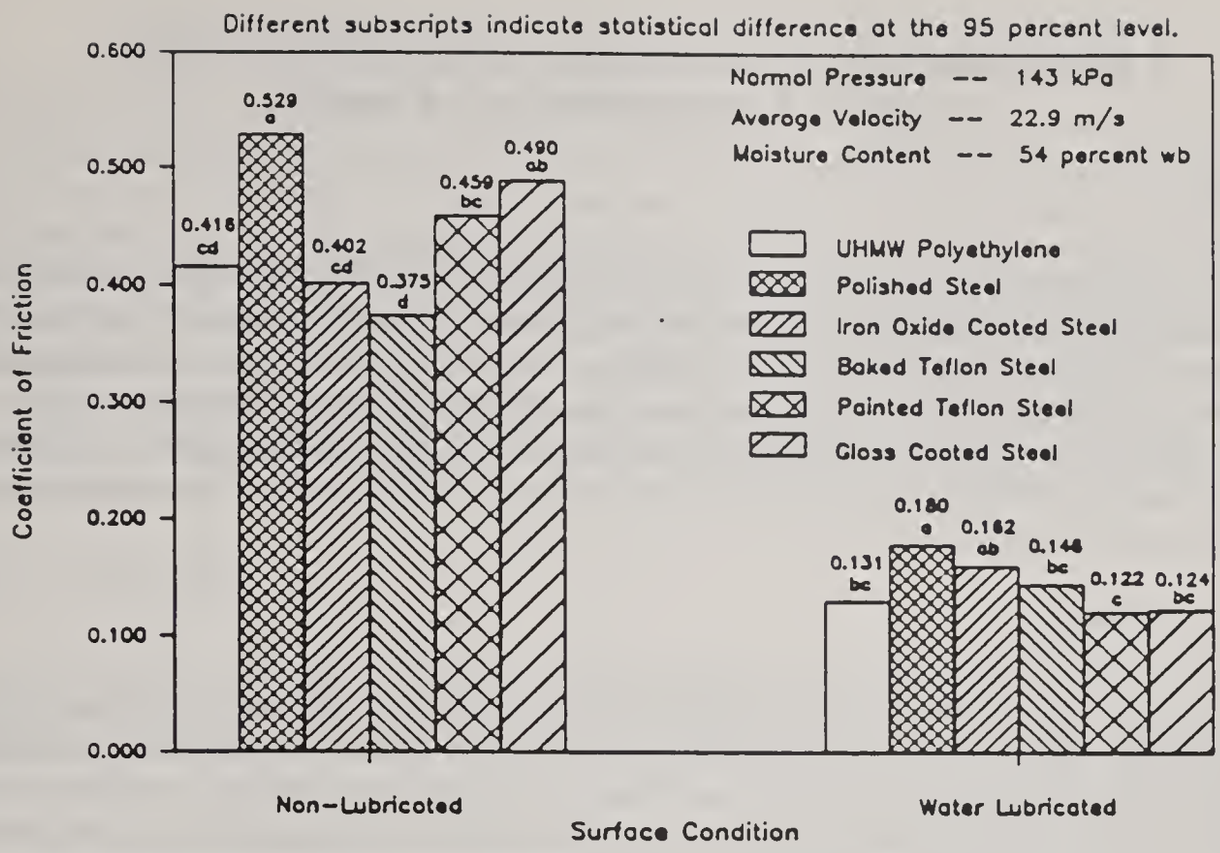


Figure 1. Coefficient of friction of alfalfa on various surfaces.

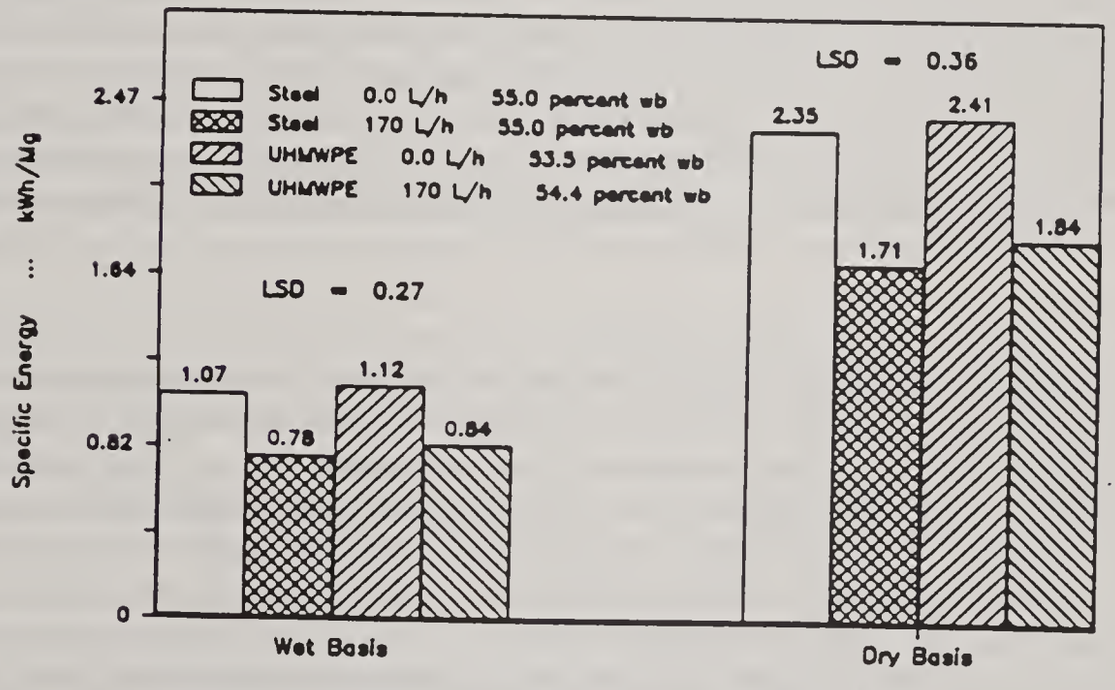


Figure 2. Effect of water lubrication rate on specific energy requirements of blower - steel and UHMW polyethylene bands.

A COMPARISON OF ALFALFA HARVEST SYSTEMS

C.A. ROTZ, D.R. BUCKMASTER and J.R. BLACK

Introduction

A computer simulation model of the dairy-forage system (DAFOSYM) has been developed to evaluate the economic consequences of management decisions and technology from a total system perspective over a wide variety of weather conditions. The model simulates the production, harvest, and utilization of alfalfa and corn to determine feed costs on representative dairy farms. The objective of this work was to use DAFOSYM to evaluate 1) the effect of baler size, 2) a comparison of rectangular- and round-bale systems, and 3) a comparison of alfalfa hay and silage systems on a typical Michigan dairy farm.

Materials and Methods

Alternative systems were compared on a representative dairy farm for 26 years of weather at East Lansing, Michigan. Parameters for DAFOSYM were set to describe a farm with 50 ha of alfalfa, 50 ha of corn, and 100 milking cows. A four-cut alfalfa harvesting system was used with first and fourth cuttings predominantly chopped and stored as silage (50 to 65% moisture). Second and third cuttings were predominantly baled as dry hay in small, rectangular bales. All alfalfa was mowed with a mower-conditioner. Silage material was field-cured in narrow swaths where the first cutting of alfalfa was not raked and fourth cutting was raked with two swaths rolled into each windrow. Hay for baling was cured in wide swaths and raked prior to baling. Harvest was begun for each of the 4 cuttings within five days of May 28, July 7, August 20, and October 15, when the crude protein of the standing crop dropped to 22%. This represented late bud to early bloom stages of maturity.

All forages were stored on the farm. Two silos were used for alfalfa where one was designated for high-quality (< 41% NDF) and the other for low- quality forage. Hay was stored in an open-front shed, also separated by quality. Corn silage and high-moisture ground ear corn were stored in top-unloaded concrete stave silos. The herd included 100 cows of milking age (26% primiparous), 30 heifers over one year old, and 36 heifers under one year old. The goal (genetic potential) for the average annual milk production of the herd was 10,000 kg/cow (3.5% fat). Prices for milk, excess feeds sold and various farm inputs were set to reflect long-term or average values relative to each other.

Several changes were made to the standard farm to compare alternatives in forage harvest and storage. An attempt was made to optimize each of the alternatives to maximize farm profit. First, rectangular balers of small, medium, and large capacity were compared. All farm parameters remained the same except rake and baler sizes. With the small baler, a single rake was used to rake the hay prior to baling. For the medium- and large-capacity balers, tandem rakes were used to roll 2 swaths into 1 windrow to improve harvest efficiency. The next alternative was to compare large round-bale systems to the standard rectangular-bale system. The rectangular baler was replaced with a medium-sized round baler. The bale wagons and bale elevator were replaced with a round-bale wagon and round-bale loader. Round bales were stored either in the storage shed or uncovered outside. The large bales were ground in a tub grinder for feeding.

The final alternatives compared to the standard system were the same farm (same land resource) with all alfalfa produced as either hay or silage. For the all hay system, only 3 cuttings were harvested because of the difficulty in harvesting hay in October. A 5% increase in average annual yield of the first 3 cuttings was modeled to represent a long-term improvement due to the less severe treatment of the crop than that which occurred with the more intense four-cut strategy. A large-sized rectangular baler was used, and the hay storage capacity was increased. Alfalfa silos were removed from the farm and a smaller forage harvester and tractor were used for harvesting corn silage. For the all silage system, the baler, bale wagons, bale elevator, and hay shed were removed from the farm. A four-cut harvest system again was used, and silo sizes were increased to contain all the forage produced.

Results and Discussion

On this 100 ha dairy farm, the high-capacity balers were more profitable than the small baler. Increasing baler size (capacity) provided a small increase in preharvest yield, i.e. larger balers provided faster harvesting which gave more time for regrowth. Harvested yield also increased with baler size and hay quality was improved. The increased fixed cost of the larger balers was more than offset by the lower operating costs due to fewer hours of operation. Labor costs also were lower with the larger balers because less labor was used at the greater harvest rate. The return above feed costs was increased about $4000/yr ($77/ha of alfalfa/yr) by using either the medium- or large-capacity balers.

Use of a large round baler caused a very small decrease in both preharvest and harvested yields. Hay quality was improved slightly due to faster harvest with the higher capacity of the round baler. Machinery costs were higher for the round bale system, but handling and feeding of large round bales required less labor which reduced labor costs. Overall, the round-bale system provided a slight improvement in farm profit of $600/yr. When the large, round bales were stored outside, forage quality and farm profit were reduced. The greater storage loss decreased the amount of excess alfalfa sold, increased the use of corn, and decreased milk production 3%. Storage costs were reduced by over $1,700 per year, but these savings did not offset the loss in milk production. Farm profit decreased by $6,000/yr ($122/ha of alfalfa/yr) when the hay was stored outside.

Either an all hay system or an all silage system were more profitable on this farm than the mixed hay and silage system. Use of the all hay system reduced both the preharvest and postharvest alfalfa yields due to the loss of the fourth cutting and the higher field losses associated with hay harvest. The need for supplemental protein decreased as the animal better utilized the forage protein in hay. Annual costs of machinery and structures decreased with the hay system because a smaller forage harvester and fewer silos were required. Milk production had a slight increase, and return for the farm was up $7,000/yr. With the all silage system, both the preharvest and post harvest yields increased substantially. Due to the higher degradability of the protein in silage, more distillers grain was needed. Machinery costs were reduced by the elimination of the baler and bale handling equipment. Labor costs were reduced, but storage costs increased because the additional silo capacity cost more than the hay shed that was removed. Milk production increased with the higher quality silage to improve farm profit by $5,800/yr.

FORAGE FIBER RECOVERY FROM DAIRY WASTE

J.Y. KIM, R.G. KOEGEL, W.P. DELZER and R.J. STRAUB

Introduction

Undigested forage fiber can be recovered from dairy manure and become a value-added product when used as bedding, for feeding, as a soil amendment, or as a biomass fuel. In addition, separation of the fibrous fraction of dairy waste facilitates the storage and handling of the remaining liquid fraction. Separation devices, available to date, have had problems which include high initial and operating costs, low throughput, and lack of reliability. The project has had two areas of emphasis during the past year: (1) design, construction and evaluation of a separation unit for the 300 cow herd at the USDFRC field facility, and (2) determination of some physical properties of various kinds of manure or waste products as an aid to separator design.

Materials and Methods

The separation unit which was designed, built and installed could be described as a double-acting, reciprocating press. The pressing chamber is annular in cross-section allowing liquid expression both inward and outward radially. The manure slurry is introduced at the middle of the press barrel; the liquid is expressed along the length of the barrel into a receiving trough, and the solids are expelled at both ends of the press alternately. The piston reciprocates through a stroke length of 23 inches at six cycles per minute.

Two pieces of apparatus were developed and used to determine the physical properties of various types of manure slurries: (1) a permeameter to determine the resistance of liquid flow at various levels of solids density and (2) a device for determining the coefficient of friction between manure solids and various machine surfaces.

Results and Discussion

The performance of the press has exceeded expectations during its first six months of operation. It has been able to process the waste from the 300 cow dairy herd in three to four hours of operation daily. When all the waste is processed it produces approximately twice the bedding requirement of the herd. The fibrous material comes from the press at approximately 70% moisture content (w.b.) and is composted for one to two weeks in one of three bins. The composting process results in the fibrous solids being pasteurized and, in addition, to reduction in the moisture content. The resulting material has good absorptive properties. It has the advantage of being produced throughout the year adjacent to the barn thus minimizing the need for stockpiling and transportation.

A "second generation" press has been designed and is currently under construction. The function of this press will be to test the versatility of the design in separating other kinds of manure as well as such materials as municipal sewage sludge and food processing waste.

Data on permeability and coefficient of friction of various manure solids have been acquired. This work will continue as an aid to press design. An example of coefficient of friction data is shown in Figure 1.

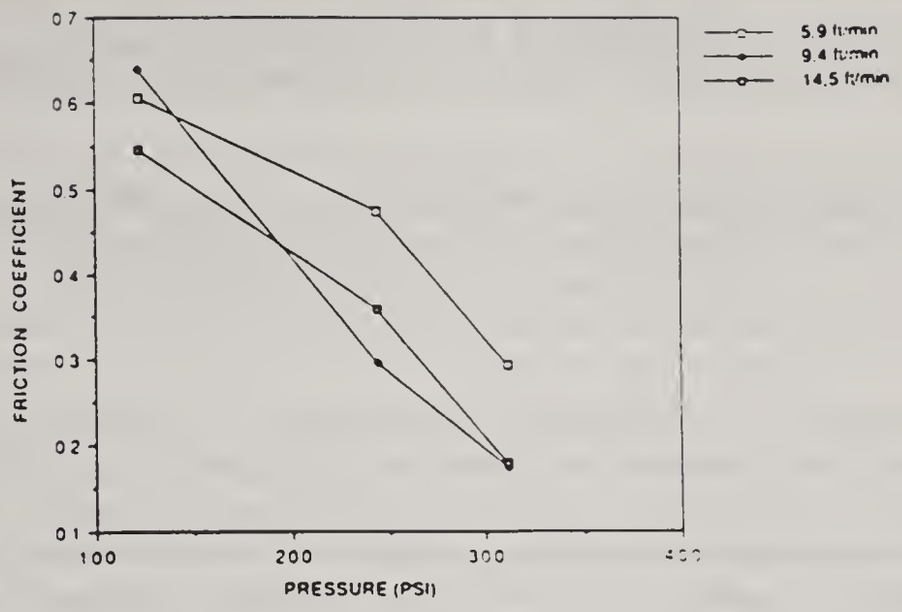
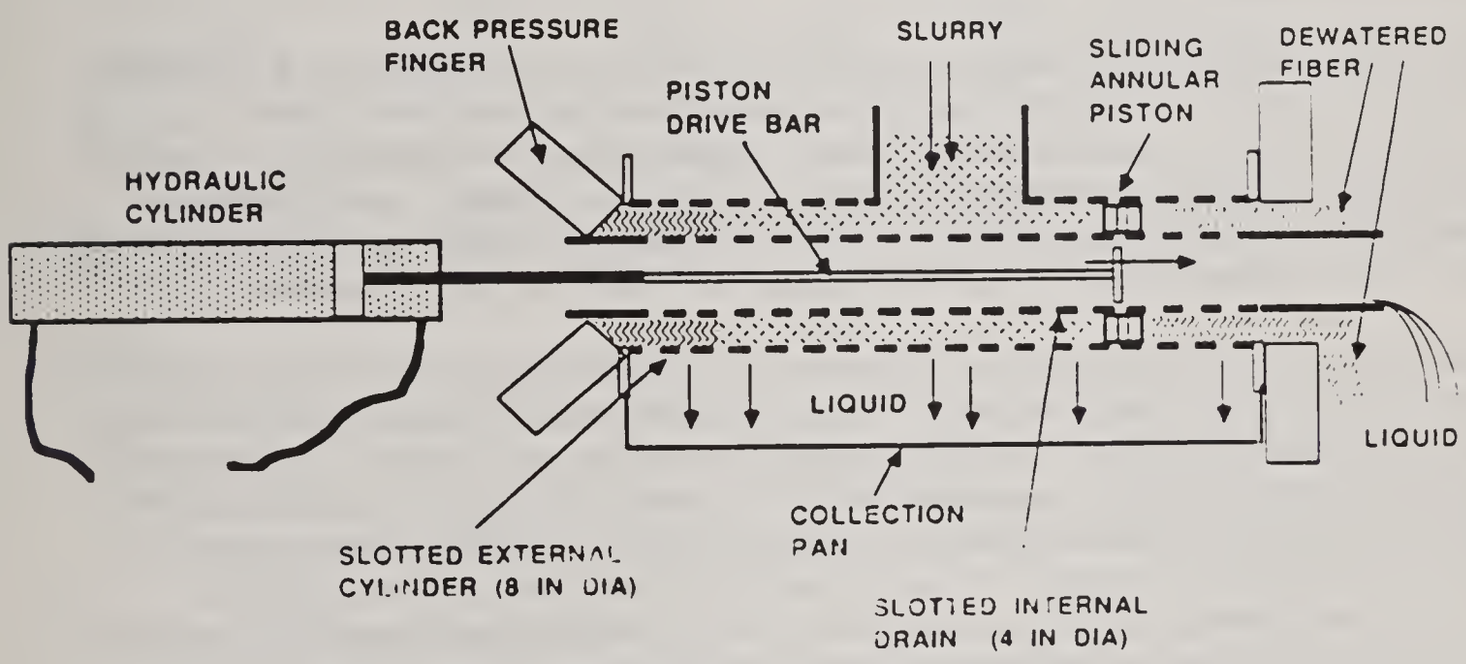


Figure 1. Friction coefficient vs pressure at maximum speed on parallel bar screen surface.



DOUBLE-ACTING ANNULAR SEPARATOR

PRESERVATION OF ALFALFA HAY WITH PROPIONIC ACID

C.A. ROTZ, R.J. DAVIS and D.R. BUCKMASTER

Introduction

Alfalfa hay is normally dried in the field to a moisture content of less than 20 percent (wet basis) to obtain proper preservation. Research data shows that under good drying conditions, 20 percent of the crop dry matter is usually lost by the time the crop is placed into storage. A 30 to 40 percent loss occurs under adverse drying conditions, and a complete crop loss can occur. The loss is directly related to the length of time the crop is in the field and inversely related to the moisture content of the crop at baling. Hay preservatives have been used to reduce this loss. Propionic acid is known to be an effective hay preservative; however, the feasibility of its use over a wide range of conditions has not been demonstrated. Our objective was to test the effectiveness of propionic acid over many years and crop conditions by measuring its effect on heating and dry matter loss during storage and visible mold, color and quality of hay following storage.

Materials and Methods

Propionic acid was applied to hay with two types of equipment. For most treatments, a commercial spray system was used. Two flood jet nozzles were located under the hood over the baler pickup to spray the hay as it moved toward the baling chamber. Liquid pressure at the nozzle was varied to provide the proper spray rate in relation to the feed rate of hay into the baler. In the first trials, propionic acid was mixed with water to form a 50% solution and the solution was applied at 20 or 40 kg/t of hay. In the remaining trials, the acid was applied at half this rate without dilution.

In several trials, an experimental injection system was used which injected the acid into a formed bale in the chamber of the baler. This system was developed to obtain a method of application that reduced the exposure of the baler to the highly corrosive acid. High-pressure liquid injection was used to insert propionic acid into a completed bale. This concept was originally developed and demonstrated in applying anhydrous ammonia. The primary component of the injector was a hydraulic cylinder called a "syringe". The syringe was directly coupled to another hydraulic cylinder which was powered by the tractor hydraulic system. Other key components included a tank, a safety shut-off valve, nozzles, solenoid valves and an electronic control circuit.

Ten field trials were conducted to evaluate propionic acid treatment of hay for improved preservation using relatively pure stands of alfalfa. All trials included propionic acid treated hay, untreated hay baled at about the same moisture content as the treated hay and untreated dry hay (about 20% moisture or less). In several trials, hay was baled both with and without the preservative treatment at two moisture levels; a portion was baled at less than 25% moisture content and the other portion was baled at greater than 25% moisture. In three trials a treatment was included where propionic acid was applied with the injection device. Hay of various treatments was baled, and ten bales of each treatment consistent in moisture content were taken to a barn for storage. Treatments were compared based upon the heat developed within a treatment stack, dry matter loss, chemical changes which indicated quality changes during storage and visual ratings of mold and color.

Results and Discussion

When compared to untreated high-moisture hay, propionic acid treatment provided a reduction in heating and dry matter loss during storage with less visible mold in the hay following storage. When compared to untreated dry hay, propionic acid treated high-moisture hay had greater heating and loss during storage and more visible mold, poorer color, and greater fiber and fiber bound protein contents following storage. The injection of propionic acid did not perform as well as spray application because the acid was not dispersed as uniformly throughout the bale.

Table 1. Comparison of untreated and propionic acid treated alfalfa hay with preservation measures averaged over similar trials and similar hay moisture contents.

Trials and treatment	Moisture content	Heat†	Storage loss	Quality from storage			Visual score*	
				CP	ADIP	ADF	mold	color
	% wb	°C-day	% DM	.....% DM.....				
Treatments ≤ 25% moisture (trials 1,4-10)								
Untreated (dry)	14.8^{a}	130^{a}	1.0^{a}	18.9^{a}	1.38^{a}	32.4^{a}	1.1^{a}	1.3^{a}
Untreated	22.8^{b}	1070^{c}	4.9^{c}	19.4^{a}	1.44^{a}	34.5^{b}	2.2^{c}	3.4^{c}
Propionic acid	23.3^{b}	413^{b}	3.5^{b}	19.2^{a}	1.45^{a}	33.1^{a}	1.5^{b}	3.1^{b}
Treatments > 25% moisture (trials 2,3,5-7,9,10)								
Untreated (dry)	14.9^{a}	139^{a}	1.2^{a}	18.8^{a}	1.14^{a}	30.0^{a}	1.0^{a}	1.2^{a}
Untreated	29.0^{b}	1782^{c}	8.2^{c}	19.8^{b}	1.46^{b}	32.2^{b}	2.8^{c}	3.7^{c}
Propionic acid	29.7^{b}	899^{b}	6.1^{b}	19.7^{b}	1.39^{b}	32.6^{b}	1.9^{b}	3.3^{b}
Treatments ≤ 25% moisture (trials 8,9,10)								
Untreated (dry)	14.7^{a}	196^{a}	1.3^{a}	17.7^{a}	1.38^{a}	34.7^{a}	1.1^{a}	1.1^{a}
Untreated	23.3^{b}	1166^{c}	5.1^{c}	18.5^{a}	1.40^{a}	38.9^{b}	2.4^{c}	3.7^{c}
Propionic (inj.)	23.0^{b}	706^{b}	3.5^{b}	18.6^{a}	1.43^{a}	37.5^{ab}	1.9^{b}	3.3^{b}
Propionic (spray)	23.0^{b}	286^{a}	3.1^{b}	18.2^{a}	1.42^{a}	36.5^{ab}	1.6^{b}	3.4^{bc}
Treatments > 25% moisture (trials 9,10)								
Untreated (dry)	15.5^{a}	261^{a}	2.1^{a}	18.4^{a}	1.30^{a}	30.9^{a}	1.1^{a}	1.0^{a}
Untreated	25.0^{b}	2067^{c}	7.2^{b}	19.9^{b}	1.43^{a}	33.0^{a}	2.3^{c}	3.7^{bc}
Propionic (inj.)	28.5^{b}	1867^{c}	7.4^{b}	19.5^{ab}	1.42^{a}	33.9^{a}	2.0^{bc}	3.7^{c}
Propionic (spray)	27.0^{b}	753^{b}	3.8^{a}	19.8^{b}	1.50^{a}	33.3^{a}	1.6^{b}	3.4^{b}

*Relative number where 1 represents green hay with no mold and 5 represents dark brown hay with heavy interior molding.

†Degree days that temperature measurements were above ambient.

^{abc}Superscript letters indicate values within an experiment which were not significantly different by Tukey's range test ($p<0.05$).

PRESERVATION OF ALFALFA HAY WITH UREA

C.A. ROTZ, J.W. THOMAS, R.J. DAVIS, M. ALLEN,
N.L. SCHULTZE PASON and C.L. BURTON

Introduction

Chemicals and other agents for preservation of hay have become popular in recent years. An effective treatment of hay at the time of baling can improve preservation to reduce storage losses and improve the quality of the forage fed to the animal. Ammonia is an effective hay preservative, but it is dangerous to handle. Urea provides another, safer method of applying ammonia. When applied to forage, urease enzyme activity in the forage breaks the urea down to ammonia and carbon dioxide to provide 0.57 units of ammonia per unit of urea. Although urea has been shown to preserve hay, a feasible method of use has not been demonstrated. This study was done to demonstrate, on a farm scale, the effect of urea treatment of high-moisture alfalfa hay on heating, dry matter loss and microbial activity during storage and visual appearance and quality of hay following storage.

Materials and Methods

Ten field trials were conducted with relatively pure stands of alfalfa to evaluate various hay treatments. All trials included urea treated hay, untreated hay baled at approximately the same moisture content as the treated hay and untreated dry hay. Urea was applied at rates of 1 to 6% of the hay dry matter. Trials 3 through 10 included propionic acid treated hay baled at the moisture content of the urea treated hay. By including these treatments, a comparison could be made between urea treatments, propionic acid treatments, and both dry and wet untreated hay. In trials 3 through 5, a treatment was included where urea treated hay was wrapped in black polyethylene to help retain an ammonia environment in the hay stack. Treatments were compared based upon the heat developed within each treatment stack, dry matter loss during storage, chemical changes which indicated quality changes during storage, visual score and mold spore count following storage.

A dry chemical applicator was developed to apply granular and powdered urea. The granular applicator was designed to satisfy two criteria: (1) accurate metering over a wide range of application rates, and (2) uniform distribution of the material throughout the bale. The metering device consisted of a small hopper, agitators, an adjustable metering orifice, a fluted wheel and two drop tubes. To obtain uniform distribution, the granular material was metered at the front of the baler and blown into the hay at the rear of the baler pickup with a fan and nozzle arrangement. A commercial spray system was used to apply propionic acid. Two flood jet nozzles were located under the hood over the baler pickup to spray the hay as it moved toward the baling chamber. Propionic acid was mixed with water to form a 50% solution and the solution was applied at 20 or 40 kg/t of hay.

Results and Discussion

Urea treatment of high-moisture alfalfa hay provided some improvement in hay preservation but the measures of improvement were not large or consistent. When compared to untreated high-moisture hay, urea treatment provided a small reduction in heating during storage with a slight improvement in hay appearance following storage (Table 1). When compared to untreated dry hay, urea treated high-moisture hay had much greater heating and loss during storage and greater fiber and fiber

bound protein contents following storage. The only consistent improvement provided by urea was an increase in crude protein content due to the nitrogen added through urea. Of the nitrogen applied, only 40% was recovered in the hay. Wrapping urea treated hay in plastic did not reduce loss or improve nitrogen retention. The plastic wrap apparently reduced the convection of heat from the stack which increased stack temperature (Table 1). Large amounts of mold were found near the plastic wrap. The small benefit obtained with the urea treatment would not justify the treatment cost.

Table 1. Average values of preservation measures for 10-bale stacks of untreated, urea treated or propionic acid treated alfalfa hay over 30 days of storage.

Trials, treatment and application rate	Heating† °C-day	Storage loss %DM	Quality from storage CP %DM	ADIP %DM	ADF %DM	Visual score*
All trials						
Untreated (dry)	189^{a}	1.5^{a}	19.0^{a}	1.82^{a}	32.4^{a}	1.1^{a}
Untreated	1112^{c}	6.3^{b}	19.3^{a}	2.03^{b}	34.4^{b}	2.9^{c}
Urea (about 3%)	948^{b}	5.8^{b}	23.2^{b}	2.06^{b}	35.2^{b}	2.6^{b}
Trials 3 to 10						
Untreated (dry)	123^{a}	1.4^{a}	19.0^{a}	1.68^{a}	31.2^{a}	1.1^{a}
Untreated	1189^{d}	5.8^{c}	19.3^{a}	1.78^{b}	32.8^{b}	2.8^{c}
Propionic acid	614^{b}	4.3^{b}	19.4^{a}	1.77^{ab}	32.3^{ab}	2.4^{b}
Urea (about 3%)	972^{c}	5.0^{bc}	23.0^{b}	1.78^{ab}	33.8^{b}	2.5^{b}
Trials 3, 4 & 5						
Untreated (dry)	76^{a}	0.5^{a}	17.5^{a}	0.97^{a}	29.8^{a}	1.1^{a}
Untreated	1344^{c}	5.1^{c}	8.3^{a}	1.06^{ab}	30.0^{ab}	3.0^{c}
Propionic acid	821^{b}	3.7^{bc}	17.9^{a}	1.17^{bc}	31.7^{ab}	2.2^{b}
Urea (about 3%)	1162^{c}	4.6^{c}	22.4^{b}	1.24^{c}	32.4^{b}	2.9^{c}
Urea (3%,wrapped)	1906^{d}	2.6^{b}	22.4^{b}	1.21^{bc}	31.3^{ab}	3.6^{d}
Trials 8, 9 & 10						
Untreated (dry)	83^{a}	1.8^{a}	21.0^{a}	2.64^{a}	32.7a	1.2^{a}
Untreated	809^{b}	6.2^{c}	20.3^{a}	2.76^{a}	35.1^{a}	2.5^{cd}
Propionic acid	625^{b}	5.9^{c}	20.8^{a}	2.59^{a}	32.7^{a}	2.8^{d}
Urea (3%)	751^{b}	5.5^{bc}	22.6^{b}	2.57^{a}	35.4^{a}	2.1^{b}
Urea (6%)	625^{b}	2.9^{ab}	24.5^{c}	2.54^{a}	33.9^{a}	2.2^{bc}

*Relative number where 1 represents green hay with no mold and 5 represents dark brown hay with heavy interior molding.
†Degree days that temperature measurements were above ambient.
^{abc}Superscript letters indicate values within an experiment which were not significantly different by Tukey's range test ($p<0.05$).

PREDICTION OF LACTIC ACID BACTERIA ON ALFALFA

R.E. MUCK and P.L. O'CONNOR

Introduction

The effectiveness of bacterial inoculants as silage additives depends on the natural level of lactic acid bacteria (LAB) on the crop at ensiling. From inoculant trials at USDFRC, inoculants appear to be economically beneficial when the inoculant population is at least ten times the natural population.

Research at USDFRC has indicated that the natural LAB population on alfalfa is affected by wilting time, temperature and rainfall, and the rate of drying in the swath. Regression equations which would allow farmers to predict LAB levels on alfalfa at ensiling have been developed and tested at USDFRC. The purpose of this study was to validate these equations at various locations in Wisconsin.

Methods

Six farms around Wisconsin (two private farms, three state experiment station farms and the USDFRC farm at Prairie du Sac) were selected for the study. Measurements on a given day at a farm were restricted to a single field. At least four samples of alfalfa were obtained from the swath prior to chopping and an additional four samples from the forage wagons as the alfalfa was emptied into a silo. LAB counts were measured on Rogosa SL agar in both sets of samples; load samples were also analyzed for moisture content. Weather data were collected from stations at the experimental farms. Records from the closest weather station were used for the private farms. Predictions of LAB on alfalfa entering a silo were based on previously developed regression equations requiring wilting time, rainfall and average air temperature during wilting and alfalfa moisture content at harvest.

Results and Discussion

Results from 1988 and 1989 are shown in Figure 1. With some notable exceptions, the regression equations predicted the actual value within an order of magnitude. Several groups of points were not predicted well. These included the first fields harvested in the year and alfalfa that was greater than 60% DM at harvest.

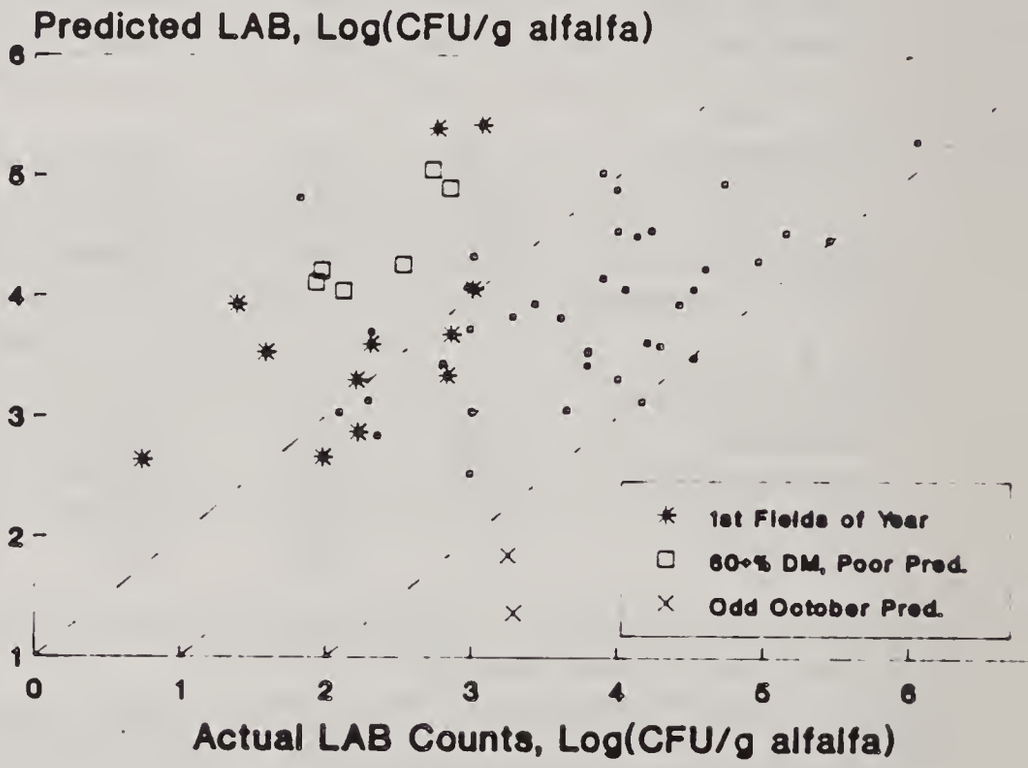


Figure 1. Summary of lactic acid bacterial count predictions on alfalfa entering silos on six Wisconsin farms, 1988-1989.

All predictions for the first fields harvested were higher than actual

values. Earlier work indicated that LAB counts on the standing crop were below detectable level and that the mowing operation provided a low level of inoculation. Perhaps inoculation from the mower is lower when first used in the spring. Also, apparent chopper inoculation is less than at other times in the year. Whatever the cause in a given sample, actual counts for these fields were never above 1000 CFU/g alfalfa.

With heavily wilted alfalfa (>60% DM), predictions were inconsistent. Some samples were predicted well whereas others were badly over-predicted. These over-predictions were not confined to a particular farm or farms. Earlier work at Prairie du Sac had found similar results so that the current results were not unexpected.

Finally, there were two fields in October from two different farms that were poorly predicted. The yields and thus the numbers of loads were low. With equipment not being used since August, forage sitting in the chopper may have caused somewhat higher counts than predicted.

Another way of analyzing the data would be to look at the number of wrong decisions a farmer would make by using the prediction equations. Eliminating the first fields of the year and the poorly predicted dry samples, there are 40 data points. Assuming one were using an inoculant supplying 10^5 CFU/g alfalfa, the cutoff for applying an inoculant would be a natural LAB level of 10^4 CFU/g alfalfa. With no error in the prediction assumed, there would have been 5 occasions when he should have applied inoculant but would not have and 5 times when an inoculant application would have been made and was not necessary. Thus, the farmer would have been right 75% of the time.

If the farmer applied inoculant when the level was at or less than 4.5 log units (0.5 unit error margin), only 2 occasions in which an inoculant would have been useful would be missed. However, the inoculant would have been used 9 times when no benefit would be expected. Milk production would be increased relative to the assumption of no error in the LAB estimate, but the average return per unit of inoculant would decline.

Overall, these regression equations appear to be reasonable predictors of natural LAB numbers on alfalfa under the climatic conditions found in Wisconsin. Combined with some economic analysis, they should be able to provide farmers with an optimum strategy for inoculant use. The applicability of these equations in other climates, however, is not known at this time.

THE BUFFERING CAPACITY OF PERMANENT PASTURE GRASS

R.E. MUCK, R.K. WILSON and P. O'KIELY

Introduction

The buffering capacity of a forage is the acid equivalent required to reduce pH from 6.0 to 4.0 per unit dry matter. The greater the buffering capacity the greater the sugar content necessary for adequate preservation of the crop via ensiling. The constituents in the plant causing buffering in this pH range are organic acids, amino acids and inorganic salts. Some of the organic acids, however, may be substrate for lactic acid bacteria. Thus, an understanding of variation in acid content as well as buffering capacity may be important for making high quality silage.

The objective of this experiment was to determine the variation in buffering capacity and plant organic acid content in grass from two permanent pastures cut under different management systems and at varying stages of maturity.

Methods

In 1988, two permanent pastures in Co. Meath, Ireland, containing perennial ryegrass and a mixture of other natural grass species, were each divided into 148 plots. Each week for 9 weeks starting May 4, four plots were randomly selected and harvested. The remaining plots were split and harvested at either May 25 (4-cut system) or June 5 (3-cut system). Regrowth in the remaining plots was sampled as in the primary growth over a 6-week period encompassing the normal cutting date. Plots not sampled still were cut at normal first regrowth dates for either the 3- or 4- cut system. This procedure continued until all the plots had been used in one of the cuttings for the harvest season which extended until late October.

Yield was measured on each plot. Samples were taken for measurement of moisture content, crude protein, fiber, and water soluble carbohydrates. Buffering capacity was determined by titration of 1.0 g of ground, dried grass (40°C) with 0.05N NaOH using a pH meter. The following acids were also analyzed in selected dried samples: oxalic, malic, malonic, succinic, citric, palmitic, oleic and stearic.

Results and Discussion

The buffering capacities of pasture grass in primary growth (Figure 1) and subsequent regrowths during the summer declined linearly with time. Among the 4 summer harvesting periods and two different pastures, the rates of decline were not statistically different with the exception of the

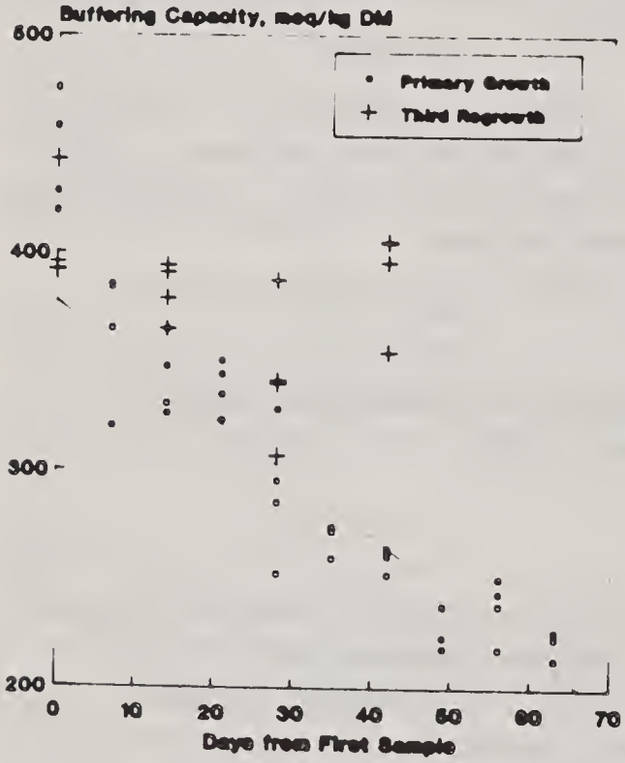


Figure 1. Buffering capacity of grass harvested from primary growth (May-June) and third regrowth (September-October) in a permanent pasture at the Grange Research Centre, Ireland.

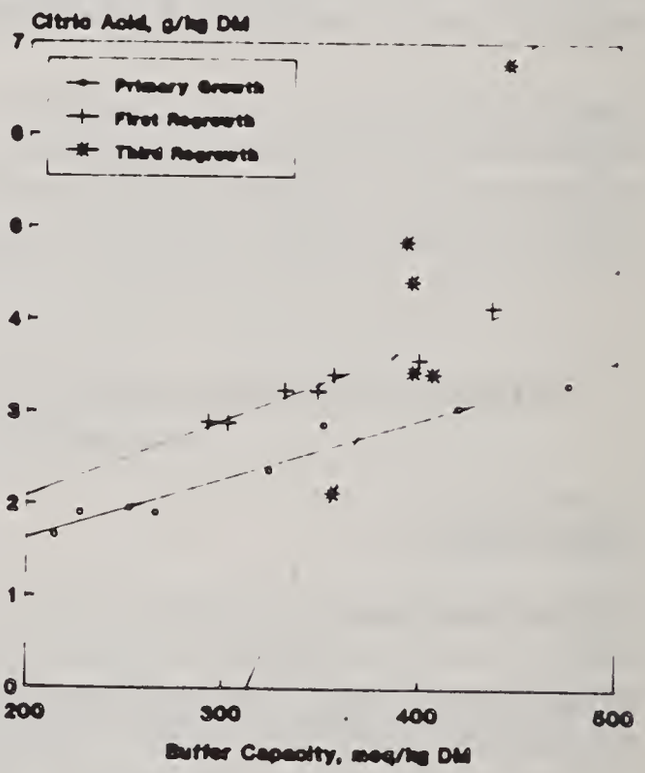


Figure 2. Citric acid content in permanent pasture grass over 1988 harvest season. Grange Research Centre, Ireland.

primary growth of one pasture. The average rate of decline was 20.0 meq/kg DM/week. These results agree with those reported for pure stands of legume and grass forages and indicate that the ensiling of less mature crops, based solely on buffering capacity, will be more difficult, particularly at high moisture contents.

The four autumn cuttings (last cut of each cutting system, two pastures) did not follow this pattern. Buffering capacities remained high and relatively constant over the period of September 7 to October 19 (Figure 1). This may be due to the slow changes in both morphological characteristics and yield in autumn.

Several of the organic acids measured (citric, succinic and palmitic) were strongly positively correlated with buffering capacity. However, the relationship was not consistent across all cuttings (See Figure 2). Variation among autumn samples was much greater than in the earlier cuttings. It appears that environmental factors affect the amount of these acids relative to buffering capacity.

The other principal acids (malic, malonic and oxalic) exhibited much poorer correlations with buffering capacity. Malic acid tended to be greatest when buffering capacity was between 300 and 400 meq/kg DM, declining above and below this range. Malonic and oxalic contents were constant across the range of buffering capacities in primary growth but increased with buffering capacity in later cuttings.

For most samples, these acids explained only 20 to 30% of the buffering capacity. This was less than half that anticipated. However, organic acids accounted for only 16% of the buffering capacity in wilted clover in a Scottish study. The results of both studies suggest that plant respiration and enzyme activity during drying and wilting may significantly alter the relative level of buffering constituents in forages.

LYSOZYME AS A POTENTIAL SILAGE ADDITIVE

R.E. MUCK, P. O'KIELY and P.L O'CONNOR

Introduction

A potential problem in ensiling wet forages (<30% DM) is a clostridial fermentation. This leads to elevated DM losses and to high butyric acid and amine contents that reduce silage intake by cattle. In the cheese industry, lysozyme has been used successfully to prevent clostridial growth during cheese ripening. The objective of this study was to determine the potential of lysozyme for preventing clostridial growth in silage.

Methods

Two trials involving lysozyme were performed in 1989. In the first trial, perennial ryegrass, which had been shaded for several days to reduce sugar content, was harvested without wilting using field equipment and ensiled in PVC silos holding 6 kg grass. Four replicates of 5 treatments (control; 85% formic acid at 3 l/ton; 20 ppm, 100 ppm and 500 ppm lysozyme) were compared. All silos were opened after 30 days and analyzed for moisture content, pH, lactic acid, volatile fatty acids, ethanol, and water soluble carbohydrates.

In the second trial, alfalfa was mowed in late afternoon, stored in a cold room overnight and chopped by hand the following morning. The alfalfa was ensiled in 100 ml test tube silos with the following 4 treatments: control, inoculation with C. tyrobutyricum at 10^5 CFU/g alfalfa, inoculum plus 20 ppm lysozyme applied separately and inoculum plus 100 ppm lysozyme applied separately. Duplicate silos of each treatment were opened at 1, 2, 4, 8, 16 and 32 days and analyzed similarly to the first trial.

Results and Discussion

The results of the first trial are shown in Table 1. A clostridial fermentation occurred in all treatments. Even at 500 ppm (25 times the effective level for control in food products), lysozyme was not effective. However, acetic acid increased and butyric acid decreased with increasing levels of lysozyme. This suggests that lysozyme did alter the dominant strains of clostridia and/or lactic acid bacteria during ensiling.

In the second trial, lysozyme was moderately successful. As shown in Figure 1, the pH of the 100 ppm lysozyme trial was similar to the control. The application of 20 ppm was unsuccessful at preventing clostridial fermentation. Assays of lysozyme activity in silage extracts indicated that there was no activity present in any of the silages after 24 h of ensiling. Thus, the effectiveness of the 100 ppm lysozyme treatment must have been from an immediate reduction in clostridial numbers rather than a continuous effect throughout fermentation.

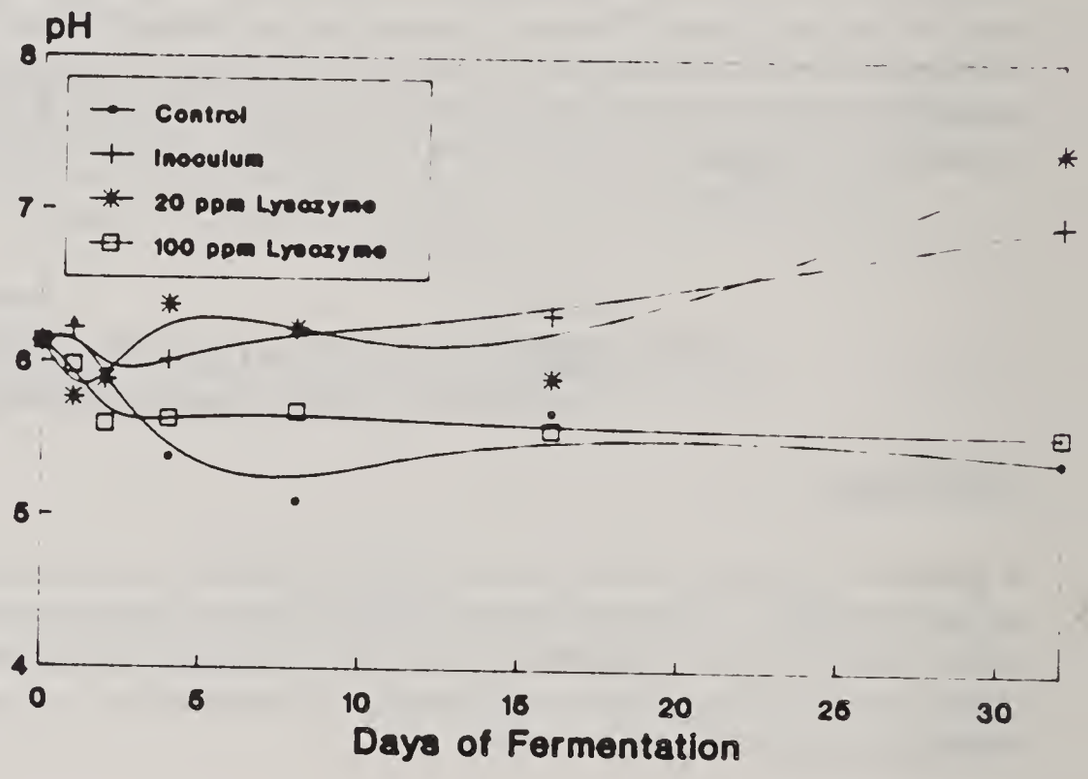


Figure 1. Alfalfa silage pH in trial 2.

Overall from these trials, it would appear that lysozyme will not be a useful silage additive. First, lysozyme would need to be effective at the 20 ppm level in order to be an economically viable additive under current conditions. Second, even though the concept did work in trial 2 with a strain that is susceptible to lysozyme, there are apparently strains in the forage environment which are not affected by lysozyme, as shown in trial 1. Considering that some strains of C. botulinum have demonstrated resistance to lysozyme, our results are not without precedent. Finally, the lysozyme assay results in trial 2 suggest that plant proteolytic activity in alfalfa may be inactivating lysozyme early in fermentation. Under such conditions, lysozyme could delay clostridial fermentation but not prevent it.

Table 1. Characteristics of perennial ryegrass silage in trial 1.

Treatment	pH	Lactic Acid	Acetic Acid	Butyric Acid
Control	5.55	0*	9.8*	9.0*
Formic Acid	5.63	5	6.9	9.4
20 ppm Lysozyme	5.50	0	10.6	8.7
100 ppm Lysozyme	5.47	0	10.9	8.5
500 ppm Lysozyme	5.47	0	11.7	7.9

*g/kg silage juice

EFFECTS OF POTASSIUM SALTS AND CITRIC ACID ON FERMENTATION OF ALFALFA SILAGE

W.L. SHOCKEY and A.L. BARTA

Introduction

Rapid attainment of low forage pH is essential for preservation of crops as silage. This is particularly true for hay crop silages that have a high protein content. Plant proteolytic enzyme activity remains active until the pH drops to about 4.5, depending upon moisture content and other factors.

Previously, researchers postulated that high concentrations of K, Ca, and Mg could increase the time required to attain low pH by forming salts and buffer systems with fermentation end products. Initial experiments with greenhouse grown alfalfa indicated that high concentrations of alkaline minerals were associated with fast rates of pH decline. Results of a second experiment in which KCl was added to alfalfa and ensiled into experimental silos indicated that the faster rate of pH decline observed in the greenhouse experiment was a result of changes in plant chemistry caused by fertilization, not alkaline mineral content per se.

A large proportion of plant K is present as potassium citrate (K- citrate). Citric acid can be fermented by lactic acid bacteria. Therefore, an experiment was conducted to compare the effects of K-citrate and citric acid to KCl addition on alfalfa silage fermentation. The hypothesis was that K addition in a form more closely representative of the naturally occurring form of K would affect the alfalfa fermentation differently than when K is added as the Cl salt.

Chopped forage was divided into four piles and no additive or KCl, K-citrate, or citric acid was added (36, 50, and 32 g/kg wet forage, respectively). The piles were mixed by hand and ensiled into 48 (6 days, 4 treatments, 2 replicates) 60 x 10 cm PVC pipes for 0, 1, 3, 7, 21 and 56 days. When silos were opened, the forage was mixed, and immediately a subsample was taken for microbiological analysis and another subsample was taken for dry matter determination. The remaining sample was frozen for subsequent lyophilization. Analyses completed were dry matter, total anaerobes, pH, nitrogen, protein nitrogen, and organic acids.

Results and Discussion

Citric acid lowered pH to 4.2 immediately after addition to forage. Microbial activity as estimated by number of anaerobes present was reduced compared to control and KCl treated forage, and proteolysis as estimated by increase in tungstic acid nonprecipitable nitrogen was only about one-half as extensive as the control or KCl treatment. Reduced proteolysis has been shown repeatedly by other workers who have lowered forage pH by acid addition.

Addition of potassium citrate affected the fermentation differently than citric acid. Final pH was higher and rate of pH decline was lower for potassium citrate treated forage compared to other treatments. Total anaerobe numbers and proteolytic activity were highest in potassium-citrate treated forage compared to other treatments. Increased proteolysis is likely the result of slow rate of pH decline. Rate of pH decline is dependent on forage buffering capacity. Historically, anionic constituents have been implicated as a major cause of forage buffering capacity. Addition of citrate, one of the major components of the organic acid fraction of alfalfa (a forage of high buffering capacity), may have increased the buffering capacity of the forage.

In contrast to previous studies in which KCl was added to wilted forage and inhibited the rate of fermentation, KCl did not affect forage pH, microbial population, or tungstic acid nonprecipitable nitrogen. Possibly the effect of salts in inhibiting microbial growth is dependent on overall osmolarity. In wilted forages osmolarity would be higher than in direct cut forage as used in this experiment. The higher moisture content of direct cut forage may have hidden any effect of added salt compared to the wilted forage of our previous experiment.

Conclusions

Citric acid immediately reduced forage pH and decreased proteolysis. Potassium citrate slowed rate of pH decline and increased proteolysis. Potassium chloride had no effect on the fermentation of direct cut alfalfa. Direct acidification is an effective way to decrease proteolytic activity in fermenting crops. Forage buffering capacity plays a very important role in controlling rate of pH decline.

Table 1. Effect of K salts and citric acid on fermentation of alfalfa silage.

Day	Control	KCl	K-citrate	Citric acid	Mean
			pH		
0	5.87	5.78	6.43	4.23	5.58
1	5.41	5.39	5.71	4.20	5.18
4	4.59	4.57	5.84	4.25	4.81
8	4.56	4.53	5.53	4.21	4.71
21	4.48	4.48	5.28	4.27	4.63
56	4.44	4.43	5.19	4.28	4.59
			LOG10 ANAEROBES/g WET FORAGE		
0	6.47	6.60	7.08	5.97	6.53
8	8.97	8.87	9.47	8.64	8.99
21	8.74	8.26	9.23	8.26	8.62
			TUNGSTIC ACID NONPRECIPITABLE NITROGEN		
			(% TOTAL N)		
0	8.0	10.1	15.6	9.0	10.7
1	19.6	16.9	14.7	11.8	15.8
4	27.2	25.1	27.0	12.8	23.0
8	31.2	28.3	29.8	16.0	26.3
21	32.5	31.2	41.0	20.9	31.4
56	35.2	31.4	44.6	21.6	33.2
			LACTIC ACID (% DM)		
0	.54	.56	.48	.50	.52
3	1.36	1.11	1.06	.85	1.09
6	1.61	1.26	1.40	1.02	1.32
28	2.22	1.82	1.82	1.32	1.79
			ACETIC ACID (% DM)		
0	5.73	5.20	5.15	4.71	5.20
3	6.42	6.41	5.71	5.60	6.03
6	6.97	6.43	6.70	6.64	6.68
28	7.08	6.07	6.89	5.42	6.36

EFFECT OF VARIOUS COMBINATIONS OF TWO LEVELS OF BACTERIAL INOCULATION AND A SUBSTRATE ADDITION ON FERMENTATION CHARACTERISTICS OF HIGH-MOISTURE ALFALFA SILAGE

C.M. WACEK and L.D. SATTER

Introduction

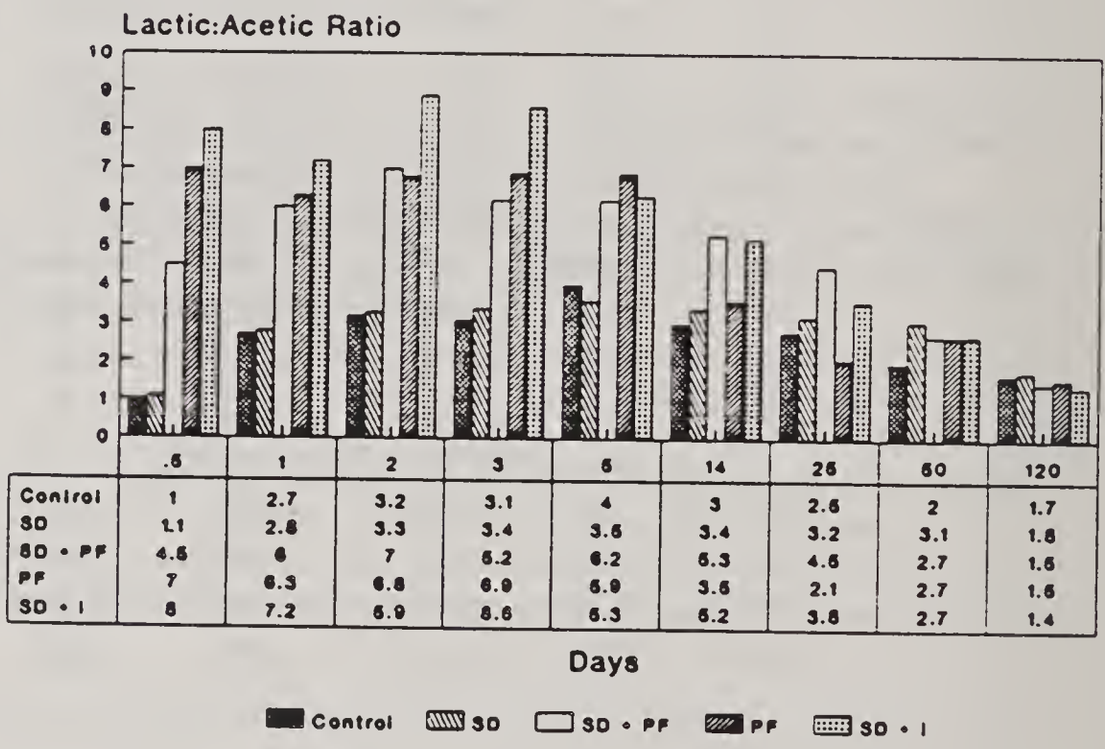
Limited substrate availability and high buffering capacity of alfalfa present formidable obstacles to successful ensiling of high mositure alfalfa. The objective of the present experiment was to determine the effect of various combinations of bacterial inoculant and substrate on fermentation characteristics of high-moisture alfalfa silage.

Materials and Methods

Third cutting alfalfa was cut and field wilted to 25 and 23% DM in trials 1 and 2. Alfalfa was chopped and ensiled in 100 ml laboratory silos. Treatments were applied as follows: 1) Control (C), no treatment added, 2) Sweet and Dry product (SD), added at 2% fresh forage weight, 3) SD and Pre-fermented inoculant (SD+PF), 4) Pre-fermented inoculant (PF), and 5) SD and inoculant (SD+I). The SD product contained rice hulls, molasses, sugar beet pulp, wheat middlings and an extruded whey product. This is an experimental product produced by Medipharm USA and is intended as a source of substrate for the silage organisms. The bacterial inoculant contained Streptococcus faecium, Lactobacillus plantarum, and Pediococcus acidolactici species and provided 7 x 10^5 lactic acid bacteria (LAB) g^{-1} chopped forage. The PF inoculant was fermented 24 hours prior to application and provided 1 x 10^7 LAB g^{-1} forage. Silos were opened at 12 hr. and days 1, 2, 3, 5, 14, 28, 60, and 120. Silage was analyzed for pH, organic acid and ethanol content, non-protein nitrogen (NPN), ammonia-nitrogen (NH_3-N) and free amino acid-nitrogen (FAA-N).

Results

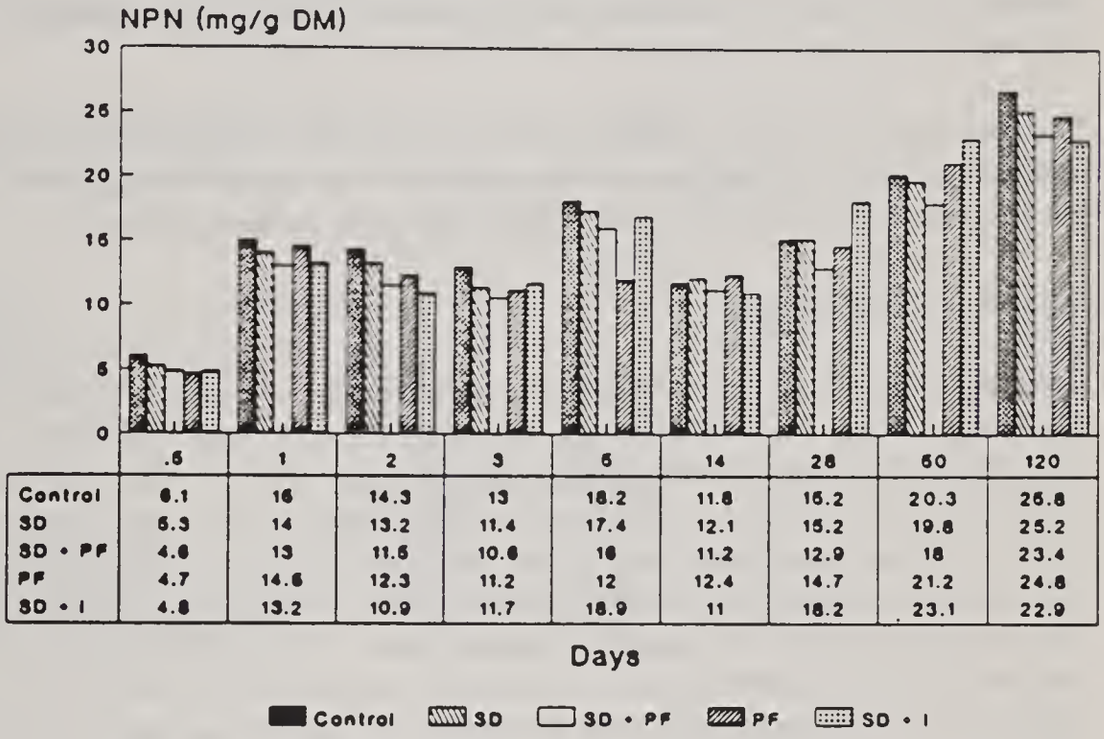
Whole plant buffering capacity averaged 500 meg/g DM and sugar content averaged 4% of alfalfa DM. Bacterial enumeration of C alfalfa revealed 8 x 10^3 LAB g^{-1} fresh forage.



	.5	1	2	3	5	14	28	60	120
Control	1	2.7	3.2	3.1	4	3	2.6	2	1.7
SD	1.1	2.8	3.3	3.4	3.6	3.4	3.2	3.1	1.8
SD • PF	4.5	6	7	6.2	6.2	6.3	4.6	2.7	1.6
PF	7	6.3	6.8	6.9	6.9	3.6	2.1	2.7	1.6
SD • I	8	7.2	6.9	6.6	6.3	6.2	3.8	2.7	1.4

Figure 1. Lactic to acetic acid ratios in silage through 120 days of ensiling.

Inoculation at both levels initially increased the rate and extent of pH decline, increased lactic acid, while restricting acetic acid, NH3-N and FAA-N production. Fermentation characteristics of C and SD were similar. However marked changes in silage composition were observed throughout the course of the experiment. While inoculation sharply increased lactic acid concentrations at 12 hours and maintained lower acetic acid concentrations that resulted in higher lactic:acetic acid ratios (Figure 1), differences diminished among treatments as fermentation progressed. Lactic content of all silages decreased and acetic acid increased with this shift in lactic and acetic acid concentrations becoming most evident at days 60 and 120. This reduction in the lactic:acetic acid ratio was most obvious for the inoculated treatments, which initially had the highest ratios.



	.5	1	2	3	6	14	28	60	120
Control	6.1	16	14.3	13	18.2	11.8	15.2	20.3	26.8
SD	5.3	14	13.2	11.4	17.4	12.1	15.2	19.8	25.2
SD • PF	4.6	13	11.5	10.6	16	11.2	12.9	18	23.4
PF	4.7	14.6	12.3	11.2	12	12.4	14.7	21.2	24.8
SD • I	4.8	13.2	10.9	11.7	16.9	11	16.2	23.1	22.9

Figure 2. Concentration of non-protein nitrogen in silages through 120 days of ensiling.

The effect of silage additives on the nitrogen fractions was less pronounced. Ammonia-N and FAA-N concentrations were lower with inoculated silages early in the fermentation, but differences between silages was very small by day 120. Non-protein nitrogen levels appeared similar in silages from days 1 through 14 (figure 2). Of interest, however, is evidence that proteolysis was not arrested in these silages. Levels of NPN continued to rise and a doubling of NPN levels occured from day 14 to 120 of ensiling.

In conclusion, although an initial trend was noted toward lower pH values, increased lactic:acetic acid ratios and decreased protein breakdown in inoculated silages, this effect diminished as the ensiling period was lengthened. The addition of the SD product had little effect, and it is possible that either a greater quantity of product or less complex substrate is required to stimulate growth of the LAB. This study points to the danger in assuming silage fermentation of high- moisture alfalfa silage remains stable after 30 or even 60 days of ensiling.

INOCULATION OF ALFALFA SILAGE WITH LARGE NUMBERS OF BACTERIA AND THE EFFECT ON LACTATION PERFORMANCE

T.R. DHIMAN, C.M. WACEK and L.D. SATTER

Introduction

Studies at the U.S. Dairy Forage Research Center suggest that there is a greater probability of animal response to feeding of inoculated silage when the treated silage contains 10 times or more the number of lactobacillus organisms as the untreated silage.

The question arises then whether a heavier inoculation rate would more likely result in increased milk production. The purpose of this experiment was to use a larger than normal inoculation rate for the silage and to evaluate utilization of the silage by lactating cows.

Materials and Methods

First crop alfalfa cut at early bloom was field wilted to 52-54% DM and ensiled as control or inoculated silage. The inoculant was supplied by Chris Hansen Company and contained Lactobacillus plantarum and Pediococcus cerevisiae species. The inoculant was applied at the rate of 3×10^5 CFU g^{-1} forage. Thirty mid lactation Holstein cows were randomly assigned to two groups of fifteen each according to their milk yield (control and treatment). The experimental design was a simple switchback with two periods of four weeks each. The diet composition and ingredient chemical composition are shown in Tables 1 and 2. Daily feed offered as a total mixed ration and feed refusal were recorded. Dairy milk yield was measured. Milk samples were collected on four consecutive milkings and analyzed for protein, fat, lactose and somatic cell counts. Body weights were measured two days in a row at the start and end of the period and once weekly during the experimental period. Feed dry matter determination was done on weekly composite samples of the total mixed ration, feed refusals and silage.

Results and Discussion

Lactation results are shown in Table 3. Feeding inoculated silage did not affect milk yield, fat percent, protein and lactose percent in the milk. Dry matter intake was slightly higher in the control group than in the treated silage.

Based on the results of the present experiment, inoculation of alfalfa silage at a higher rate did not affect animal performance. Cows in the study were in midlactation and may not have been as responsive as cows in early lactation would have been.

Table 1. Ingredient composition of diet.

Ingredient	Control	Treatment
	(% dry basis)	
Control silage	65	—
Inoculated silage	—	65
Shelled corn	33.2	33.2
Dicalcium phosphate	1.1	1.1
Trace mineralized salt	0.7	0.7
Vitamin ADE	trace	trace

Table 2. Chemical composition of feed ingredients (%).

	Silage		Shelled Corn
	Control	Inoculated	
Dry matter	51.7	54.4	89.7
Crude protein	18.7	18.9	9.2
NDF	35.4	38.2	—
ADF	30.8	30.9	—
pH	4.67	4.51	—
NPN, % of total-N	52.9	50.3	—
Free amino acid N, % of total-N	19.2	18.6	—
Ammonia N, % of total-N	5.0	4.7	—

Table 3. Effect of silage inoculation on lactation performance.

	Control Silage	Treated Silage
Milk yield, kg/d	26.5	26.3
Fat, %	3.15	3.16
Protein, %	2.95	2.96
Lactose, %	4.84	4.83
Somatic cell counts	200	227
Dry matter intake, kg/d	19.4	19.1
Ave. body wt. loss during experimental period, kg/8 weeks	-1.0	-.8

No significant difference for any parameter ($P<.05$).

EFFECT OF HIGH INOCULATION RATE OF ALFALFA SILAGE ON SILAGE FERMENTATION AND ANIMAL PERFORMANCE

C.M. WACEK and L.D. SATTER

Introduction

Previous research at the USDFRC suggests that the potential for benefit from silage inoculants is much higher if the inoculated silage has at least ten times the number of lactic acid bacteria (LAB) as the untreated silage. One way to achieve a high ratio of LAB in treated relative to untreated silage is to apply larger numbers of bacteria. The objective of this experiment was to add 3 to 5 times the usual number of inoculant bacteria, thus improving chances of benefit from the inoculation.

Materials and Methods

First crop alfalfa was cut at early bloom, field wilted to 59% DM and ensiled in three concrete stave silos. Treatments consisted of 1) control silage (C), 2) Medipharm inoculant supplying 5 x 10^5 LAB g-1 forage (M) and 3) Chris Hansen inoculant supplying 3 x 10^5 LAB g^{-1} forage (CH). Medipharm inoculant contained Streptococcus faecium, Lactobacillus plantarum and Pediococcus acidolactici species. The Chris Hansen inoculant contained Lactobacillus plantarum and Pediococcus cerevisiae species. Increased numbers of M inoculant were achieved by pre-fermenting the inoculant for 24 hours.

Core samples were removed through sealed holes in silo doors at days 1, 2, 4, 8, and 28. Core samples and samples collected during feedout were analyzed for organic acids and ethanol, non-protein nitrogen (NPN), ammonia-nitrogen (NH_3-N) and free amino acid-nitrogen (FAA-N).

Sixty-six holstein heifers were divided into groups of 33 light and 33 heavy heifers. Animals were ranked based on weight and randomly assigned to one of six pens. Initial heifer weights, for light and heavy pens, averaged 267 and 378 kg. Diets were fed once daily to provide a 5 to 10% refusal rate and consisted of 98% alfalfa silage plus mineral and vitamin supplement.

Results

Buffer capacity and sugar content of the alfalfa averaged 512 meg/kg and 68 mg/g DM. Numbers of LAB on the C, CH and M silages going into the silo were 2 x 10^3, 3 x 10^5 and 5 x 10^5 LAB^{-1} forage. Higher bacterial numbers increased the rate of pH decline past day 1 and 2 for CH and M inoculants, however pH values were similar by day 8 for treated and C silages. Organic acid content of silage at feedout are in Table 1. No significant differences were noted between treatments with the exception of a slightly lower production of propionic acid for CH silage. Inoculation did not reduce NPN, NH_3-N and FAA-N concentrations relative to C silage (Table 1). Total DM recovery from C, CH and M silos averaged 91, 95 and 94%.

Average daily DM intakes, weight gains and feed efficiencies are presented in table 2. No significant differences were noted for any parameter ($P<.05$) for light or heavy heifers, although a tendency was noted for greater feed efficiencies with CH inoculant. Greater weight gains and higher feed efficiencies were achieved for light relative to heavy heifers.

It was expected that the higher inoculation rates employed would result in the inoculant bacteria overwhelming the epiphytic bacteria, resulting in improved silage and possibly improved animal performance. The reasons for this not occurring are not known.

Table 1. Influence of bacterial characteristics of alfalfa silage as measured at feed out.

Parameter	Control (C)	Chris Hansen (CH)	Medipharm (M)	SD[4]
Lactic acid, mg/g DM	56.4	58.8	61.2	6.23
Acetic acid, mg,g DM	7.0	7.6	8.6	1.33
Succinic acid, mg/g DM	2.5	2.2	2.2	.54
Propionic acid, mg/g DM	5.8[A]	5.2[B]	5.9[A]	.40
Ethanol, mg/g DM	1.7	1.4	.8	.77
NPN, % of total N[2]	52.9	50.3	54.5	—
NH_3-N, % of total N	5.0[A,B]	4.7[B]	5.3[A]	—
FAA-N, % of total N	19.2	18.6	19.8	—

[1]NPN=Non protein nitrogen
[2]N=Nitrogen
[3]NH_3-N=Ammonia nitrogen
[ABC]Means within rows with unlike superscripts differ significantly at P<.05.
[4]Standard deviation

Table 2. Average daily dry matter intake and weight gain of Holstein heifers.

	Control (C)	Chris Hansen (CH)	Medipharm (M)
Weight gain, kg/d[1]			
Light heifers	1.13	1.09	1.13
Heavy heifers	.86	.92	.82
Dry matter intake, kg/d			
Light heifers	8.48	7.96	8.47
Heavy heifers	9.48	9.30	8.81
Efficiency, kg feed/kg gain			
Light heifers	7.50	7.30	7.50
Heavy heifers	11.0	10.1	10.7

[1]No significance for any parameters at P<.05.

EFFECT OF CELLULOSE FINE STRUCTURE ON ITS IN VITRO DIGESTION KINETICS BY MIXED RUMEN MICROFLORA

P.J. WEIMER, J.M. LOPEZ-GUISA and A.D. FRENCH

Introduction

Cellulose is the primary component of forage cell walls and thus a major contributor to ruminant nutrition. Although it has a very simple primary structure, cellulose has a complex and variable higher order structure. Little is known of the relationship between this "fine structure" and the digestion of cellulose, although one fine structural feature—crystallinity— is generally regarded as an important determinant of digestion rate. In order to examine cellulose structure/digestion relationships more completely, we obtained or prepared a variety of different celluloses, measured several fine structural parameters, and determined the digestion kinetics of the celluloses by mixed rumen microflora in vitro.

Materials and Methods

Eight commercial celluloses were obtained for this study. One of the celluloses (Sigmacell 50, or SC50) was modified by treatment with different concentrations of phosphoric acid (72-80%, 0°C, 1 hr) to yield six different phosphoric acid-swollen celluloses. Some SC50 was also mercerized in 20% NaOH for 1 hr at 0°C. X-ray diffraction was used to determine both crystallinity (i.e., degree of interchain hydrogen bonding to form a rigid matrix) and allomorphic form (i.e., the arrangement of individual atoms within the unit cell of the crystallites). Crystallinity was also measured by acid hydrolysis kinetics. Gross specific surface area (GSSA , i.e., the surface area per unit mass which was accessible to whole microbial cells but which excluded the vastly larger surface area due to pits, capillaries, and micropores within the cellulose fibers) was determined by microscopic measurement of the physical dimensions of 300 random cellulose particles; these dimensions were fitted to geometric mensuration formulae based on the assumption that the cellulose fiber was either shaped as a cylinder or as a rectangular parallelpiped; both models yielded identical GSSA values if the latter model assumes an aspect ratio (width/depth) of unity.

In vitro digestion experiments were conducted under CO_2 in serum vials that contained 1% cellulose in reduced McDougall buffer, inoculated with 10% (v/v) of rumen fluid. Vials were shaken at 100 rpm and 39°C to eliminate the confounding effects of differential settling rates of the different celluloses. Residual cellulose was recovered by a modified detergent procedure. Weight-loss data were fitted to a discontinuous first-order kinetic equation that included a rate constant and a discrete lag time. These kinetic parameters were normalized to those for untreated SC50 to permit comparison of data from different experiments using different fresh rumen fluid inocula.

Results and Discussion

All of the celluloses were completely digested within 48 h, following lag periods of 8-19 h. The commercial celluloses were all of the natural type I allomorph, and showed considerable

variations in crystallinity and GSSA (Table 1). Comparison of a subset of four isocrystalline nonaggregating celluloses revealed a strong correlation between the digestion rate constant and GSSA (r= +0.99), and a weaker negative correlation between lag time and GSSA (r=-0.96). Crystallinity appeared to be relatively unimportant in determining digestion kinetics, since celluloses of different crystallinity but different similar GSSA were digested at similar rates.

Treatment of SC50 with H_3PO_4 resulted in a series of less-crystalline swollen celluloses (Table 2). Treatment at the lower concentrations (<77% H_3PO_4) resulted in a slight enhancement of digestion rate, but more aggressive treatment resulted in a more slowly-fermented material, despite extensive swelling and decrystallization. This reduced fermentation rate was apparently due to a partial conversion from the natural (type I) to the type II allomorph, with a resultant change in the dimesions of the unit cell. Indeed, complete conversion of SC50 to the type II allomorph by alkaline mercerization resulted in a halving of the fermentation rate and doubling of the lag time.

The strong dependence of kinetic parameters on GSSA observed in the type I celluloses, along with the failure of phosphoric acid-swelling to improve the cellulose fermentation, suggest that the cellulolytic enzymes of rumen microbes are cell-bound rather than extracellular. The dramatically decreased fermentation rate and increased lag time following conversion of native cellulose to its type II allomorph suggest that rumen microbes, in contrast to aerobic cellulolytic fungi (the classical model system for the study of cellulolysis), respond slowly and relatively ineffectively to small changes in the dimensions of the cellulose unit cell. This has important implications for those biomass pretreatments which cause allomorphic conversion of cellulose.

Table 1. Fine structure and digestion parameters for commercial Type I celluloses.

Cellulose	RCI (ah)[a]	RCI (xrd)[b]	GSSA[c] (m^2/g)	normalized rate[d] Expt 1	normalized rate[d] Expt 2	normalized lag[e] Expt 1	normalized lag[e] Expt 2
SC50	90.1	94.9	ND[f]	1.00 (0.01)	1.00 (0.03)	1.00 (0.01)	1.00 (0.05)
PH101	89.3	97.5	ND[f]	1.04 (0.07)	1.11 (0.01)	1.03 (0.04)	1.00 (0.06)
SC20	86.3	96.6	0.301	0.95 (0.09)	0.93 (0.01)	0.85 (0.10)	0.78 (0.03)
SC100	64.8	78.9	0.310	0.95 (0.05)	0.97 (0.12)	1.03 (0.04)	1.15 (0.07)
alpha	72.4	92.2	0.177	0.81 (0.01)	0.88 (0.02)	1.13 (0.00)	1.29 (0.03)
CC31	90.8	98.4	0.228	0.63 (0.01)	0.73 (0.01)	1.06 (0.00)	1.20 (0.00)
CF11	88.2	99.4	0.167	0.53 (0.02)	0.60 (0.00)	1.21 (0.05)	1.33 (0.00)
CF1	87.1	97.4	0.158	0.47 (0.01)	0.54 (0.02)	1.18 (0.01)	1.26 (0.03)

[a]Relative crystallinity index as determined by acid hydrolysis kinetics.
[b]Relative crystallinity index as determined by x-ray diffraction.
[c]Gross specific surface area estimated by microscopy
[d]Rate constant for cellulose / rate constant for SC50; the rate constants for SC50 inExpts 1 and 2 were 0.0859 and 0.0596 h^{-1}, respectively. Coefficients of variation in parentheses.
[e]Lag time for cellulose / lag time for SC50; the lag times for SC50 in Expts 1 and 2 were 9.76 and 8.32 h, respectively. Coefficients of variation in parentheses.
[f]Not determinable due to aggregation of particles.

Table 2. Fine structure and digestion parameters of treated SC50 celluloses.

Treatment	Extent of allomorphic conversion	RCI	normalized rate	normalized lag time
None	None	94.9	1.00 (0.02)	1.00 (0.06)
72.0 % H_3PO_4	None	96.7	1.03 (0.07)	0.91 (0.08)
75.5 % H_3PO_4	None	97.1	1.16 (0.05)	1.07 (0.06)
77.0 % H_3PO_4	Slight	82.2	1.08 (0.08)	1.18 (0.06)
78.0 % H_3PO_4	Extensive	77.1	0.85 (0.01)	1.24 (0.02)
80.0 % H_3PO_4	Extensive	54.2	0.88 (0.05)	1.34 (0.06)
20 % NaOH	Complete	~ 80	0.57 (0.01)	1.92 (0.01)

ATTACHMENT OF *FIBROBACTER SUCCINOGENES* TO CELLULOSE AND CELLULOSE DERIVATIVES

P.J. WEIMER and J.K. SCHMIDT

Introduction

Attachment of fibrolytic ruminal bacteria to forage cell wall is a prerequisite to rapid fermentation of their polysaccharide components. Relatively little is known about the factors controlling the rate or extent of this attachment. In order to obtain a better understanding of the attachment process, we examined the attachment of *Fibrobacter succinogenes*, a predominant ruminal celluloytic bacterium, to various pure celluloses and cellulose ethers (cellulose analogs in which hydroxyl functions were replaced by charged or uncharged ether groups of varying atomic size).

Materials and Methods

Fibrobacter succinogenes strain S85 was usually grown to mid-exponential phase (18-24 h, although longer growth periods were also tested) under a CO_2 atmosphere, in serum vials that contained a modified Dehority medium supplemented with 0.1% (w/v) of Avicel PH101 microcrystalline cellulose. Cellulose-free cell suspensions for binding studies were collected by filtration through 5 µm polycarbonate membranes. The cell suspensions were added to 8 mL serum vials that contained 3 mL of reduced McDougall buffer plus 100 mg of substrate under a CO_2 atmosphere. The substrates consisted of various commercial celluloses and cellulose ethers. The vials were shaken at 150 rpm in a 39°C bath for 10-60 min, and their contents vacuum filtered through 5 µm polycarbonate membranes. Filtrates and the material retained on the filter were separately alkaline-digested and analyzed for protein by the Bradford method. Controls included cell-free culture suspensions (to account for extracellular protein in the cell suspension), and culture suspensions lacking substrate (to permit estimation of total protein recovery).

Results and Discussion

Attachment of *F. succinogenes* S85 to cellulose (Avicel PH101) was maximal within 10 min of exposure, and was strongly influenced by culture age (Table 1). Extent of attachment to the substrates exhibited the general pattern: cationic cellulose ethers>cellulose>anionic cellulose ethers, suggesting that attachment is partially dependent upon electronic interactions at the bacterial surface

(Table 2). Extent of attachment of cells to pure cellulose was independent of its particle size and crystallinity, suggesting that widely-reported variations in fermentation kinetics of different celluloses are not due to variations in attachment during the lag phase prior to the initiation of fermentation.

Attachment of cells to cellulose was inhibited by soluble neutral cellulose ethers (e.g., methylcellulose), and by hydrolyzates of cationic cellulose ethers, suggesting that these substrates may be useful for removing adherent bacteria from forage materials to determine the relative fraction of attached and unattached microbial cells in digesta samples.

Table 1. Effects of culture age on attachment of *F. succinogenes* S85 to Avicel PH101.

Age	Growth phase	% Attachment of	
		Cell protein	Supernate protein
1 day	Mid-exponential	40.0	6.6
2 day	Late exponential	25.4	1.7
6 day	Late stationary	6.7	17.5

Table 2. Attachment of *F. succinogenes* S85 to different celluloses and cellulose ethers.

Expt.	Substrate	Type	% Attachment
I	DEAE Cellulose	Strongly cationic cellulose ether	72.7[a]
	ECTEOLA Cellulose	Weakly cationic cellulose ether	69.4[a]
	Avicel PH101	Microcrystalline cellulose	56.1[b]
	CM Cellulose	Strongly anionic cellulose ether	9.9[c]
II	Avicel PH101	Coarse microcrystalline cellulose	37.3[a]
	Sigmacell 20	Fine microcrystalline cellulose	43.6[a]
	Whatman CF-1	Long fibrous cellulose	33.1[a]

[a,b,c]Substrates in same experiment having different superscripts differ ($P<.05$).

ENERGY SPILLING REACTIONS IN RUMINAL BACTERIA

J.B. RUSSELL and H.J. STROBEL

Introduction

It had generally been assumed that microbial growth yields in the rumen were fairly constant, but it is now apparent that yield can vary significantly. The rumen evolved as an energy-limited habitat which was primarily dependent on herbage, but modern feeding practices now emphasize the use of cereal grains. These cereal based diets are usually correlated with a depression in rumen microbial yield. Because microbial protein is the primary amino acid source of ruminant animals, there is often a positive relationship between growth yields and animal production.

Energy yielding pathways in bacteria are not always tightly coupled to growth, and experiments with non-rumen bacteria demonstrated that energy-sufficient cultures can continue to metabolize energy sources in the absence of growth. The terms energy spilling, overflow metabolism, slip reactions, and uncoupling have all been used to describe this phenomenon, but the process was not well understood.

Streptococcus bovis is a rapidly growing bacterium that flourishes in the rumen when animals are fed large amounts of cereal grains, and it is responsible for the onset of ruminal acidosis. S. bovis can be inhibited by monensin, an ionophore which is commonly used as a feed additive in beef cattle rations.

Materials and Methods

Batch cultures of the ruminal bacterium, Streptococcus bovis, were inhibited with chloramphenicol or nitrogen deprivation and bacterial heat production was measured with a sensitive microcalorimeter.

Results

Streptococcus bovis had a doubling time of approximately 21 min and the rate of bacterial heat production was proportional to the optical density (1.72 μW/μg protein). If exponentially growing cultures were treated with chloramphenicol, there was a decline in heat production, but the rate was greater than 0.30 μW/μg protein even after growth ceased. Since there was no heat production after glucose depletion, this growth-independent energy dissipation (spilling) was not simply due to endogenous metabolism. Stationary cells which were washed and incubated in nitrogen-free medium containing an excess of glucose produced heat at rate of 0.17 μW/μg protein. Monensin and tetrachlorosalicylanilide (TCS), compounds which facilitate an influx of protons, caused a more than 2-fold increase in heat production. Dicyclohexylcarbodiimide (DCCD), an inhibitor of the F_1F_0 proton ATPase, virtually eliminated growth-independent heat production regardless of the mode of growth inhibition. Because DCCD had little effect on the glucose transport, it appeared that the combined action of proton influx and the membrane bound ATPase was responsible for energy spilling.

Discussion

The impact of energy spilling reactions on rumen microbial yields had never been fully assessed. It has been estimated that approximatley 4-10% of the feed energy in ruminants was dissipated as heat of fermentation, but these estimates were based on indirect calorimetry rather than direct measurments. Walker and Forrest (1964) used an adiabatic calorimeter to measure the heat production of mixed rumen bacteria, but the relationship between growth and heat production was not studied. Based on a total ruminal bacterial mass of 0.8 kg protein and a glucose consumption of 6.9 mmol h^{-1} g $protein^{-1}$, energy spilling could account for as much as 5.5 mol glucose/h or 1 kg/h.

While it is unlikely that all rumen bacteria can spill energy as quickly as S. bovis, preliminary experiments indicated that nitrogen-limited, mixed rumen bacteria were able to consume glucose at 1/4 this rate. Whether energy spilling occurs to a significant extent in the rumen has yet to be determined, but one can envision circumstances when growth might be either too rapid or limited by nutrients other than energy. Practical experience has indicated that mixed rations and frequent feeding can enhance animal productivity, and these improvements might be related to a better balance of nutrients throughout the feeding cycle. The potential impact of proton cycling in the ruminal bacteria is illustrated by the observation that mixed ruminal bacteria had a 50% lower growth yield when the pH was decreased from 6.7 to 5.7 (Strobel and Russell, 1986).

While energy spilling reactions may be wasteful from the perspective of animal production, they may be advantageous to microbes in natural environments like the rumen. When growth is energy limited, the maintenance of high transport activity is beneficial. However, if the supply of energy sources increases abruptly, transport activity can transiently exceed the growth capacity. Energy spilling reactions offer a means of protecting the cells from potentially toxic concentrations of metabolic intermediates.

THE INVOLVEMENT OF SODIUM IN THE TRANSPORT ACTIVITIES OF RUMINAL BACTERIA

J.B. RUSSELL

Introduction

The rumen contains an abundance of sodium (usually 100 mM) and it has been described as an "inland sea". Many rumen bacteria have an absolute requirement for sodium, but the nature of this requirement had never been defined. Monensin and lasalocid are routinely fed to beef cattle, and these ionophores can destroy the sodium gradients across the cell membranes of ruminal bacteria.

Materials and Methods

Washed ruminal bacteria were energized with glucose or treated with valinomycin, loaded with potassium and diluted into potassium or sodium or potassium containing buffers to create a membrane potential and/or sodium gradients across cell membranes. The uptake of carbon-14 labeled amino acids was monitored from 0 to 120 seconds.

Results and Discussion

Streptococcus bovis, a ruminal bacterium which is involved in the onset of ruminal acidosis, transported serine, threonine or alanine, but only when the cells were incubated in sodium buffers. If glucose-energized cells were washed in potassium phosphate and resuspended in potassium phosphate buffer, there was no detectable uptake. Cells de-energized with 2-deoxy-glucose and incubated in sodium phosphate buffer were still able to transport serine, and this result indicated that the chemical sodium gradient was capable of driving transport. However, when the de-energized cells were treated with valinomycin and diluted into sodium phosphate to create both an artificial membrane potential and a chemical sodium gradient, rates of serine uptake were 5 fold greater than in cells having only a sodium gradient. If de-energized cells were pre-loaded with sodium (no membrane potential or sodium gradient), there was little serine transport. Monensin strongly inhibited sodium-dependent uptake of the three amino acids. Membrane vesicles loaded with potassium and diluted into either lithium or choline chloride were unable to transport serine, but rapid uptake was evident if sodium chloride was added to the assay mixture. Serine transport had an extremely poor affinity for sodium and more than 30 mM was needed for half maximal rates of uptake. Serine transport was inhibited by an excess of threonine, but an excess of alanine had little effect. Results indicated that S. bovis had separate sodium symport systems for serine/threonine and alanine, and either the membrane potential or chemical sodium gradient could drive uptake.

A recently isolated ruminal peptostreptococcus which produced large amounts of branched-chain volatile fatty acids grew rapidly with leucine as an energy source in the presence but not the absence of sodium. Leucine transport could be driven by an artificial membrane potential only when sodium was available, and a chemical gradient of sodium also drove uptake. Because $sodium^{+}$ was taken up with leucine and a proton gradient could not serve as a driving force (with or without sodium), it appeared that leucine was transported in symport with sodium. The leucine carrier could use lithium as well as sodium and had a single binding site for sodium. The affinity constant for sodium was 5.2 mM, and affinity constant and maximum velocity for leucine were 77 uM and 328 nmol/mg protein/min, respectively. Since valine and isoleucine competitively inhibited (inhibitor constant of 90 and 49 uM, respectively) leucine transport, it appeared that the peptostreptococcus used a common carrier for branched-chain amino acids. Valine or isoleucine were taken up rapidly, but little ammonia was produced if they were provided individually. The lack of ammonia could be explained by an accumulation of reducing equivalents. The ionophore, monensin, inhibited growth, but leucine was taken up and deaminated at a slow rate. Monensin caused a loss of potassium, an increase in sodium, and a decrease in intracellular pH. The inhibition of growth was consistent with a large decrease in ATP. The capacity of the ruminal peptostreptococcus to transport and deaminate branched-chain amino acids and its sensitivity to monensin clarifies the observation that monensin decreases branched-chain volatile fatty acid production in vivo. Branched chain volatile fatty acids are required by cellulolytic ruminal bacteria.

For many years it was assumed that active transport in bacteria was driven by ATP hydrolysis or proton gradients, but it is now evident that a variety of bacteria including ruminal bacteria have transport mechanisms which are coupled to sodium symport.

THE EFFECT OF HEAT TREATMENT ON THE ESTIMATED RUMINAL ESCAPE OF PROTEIN IN ALFALFA HAY

J.H. YANG, G.A. BRODERICK, R.G. KOEGEL and D.B. RICKER

Introduction

Alfalfa hay is an important feed ingredient for dairy cattle in the United States. However, studies have shown that cows fed alfalfa hay or silage produced less milk protein and milk with lower protein content due to the high rumen degradability of alfalfa protein.

Heat treatment of oil seeds has enhanced protein resistance to ruminal degradation and improved protein utilization. But there is little evidence in the literature to show if ruminal escape of alfalfa protein also can be increased by heat treatment. Our objectives were to study the effect of heat treatment on the ruminal degradability of protein in alfalfa hay and to identify an optimal method of heat treatment.

Materials and Methods

Two kinds of alfalfa hay, conventional and shredded at time of mowing, were heated in either a forced air oven or a steam pressure cooker at different temperatures for different lengths of time. Both hays were harvested at first cutting (June 15, 1988) from the same field at the same stage of maturity. The shredded hay was made using a macerator normally used to achieve a high rate of field drying. Both hays were chopped to about 7.5 cm in length.

About 120 g of each alfalfa hay was put in a tray (70 X 25 X 5 cm) with a wooden frame and wire screen at the top and bottom. One layer of cheesecloth was spread on the bottom to keep the small particles of hay from falling through the screen during heating. The hay heated in the pressure cooker also was wrapped with one layer of cheesecloth.

Heated hays were ground through a 1 mm screen and analyzed for DM, total N and ADIN. Ruminal degradation rates of protein were measured by an inhibitor in vitro system and extents of ruminal escape were estimated assuming a ruminal passage rate of .06/hr (Broderick, Brit. J. Nutr. 58:463, 1987). Estimated protein escapes from the rumen were corrected for indigestible N based on ADIN content.

Results and Discussion

Estimated net protein escape (i.e., total protein escape minus ADIN) of both conventional and shredded hays was increased by the oven or steam heating (Figures 1 and 2). The temperatures used in oven heating were higher, but so were net protein escapes. Optimal treatment times, as indicated by the greatest increase in net protein escape for oven heated hays in this trial were 120 min at 140°C, 60 min at 150°C, and 30 min at 160°C. The same optimum heating times were found for heat

treatment of full fat soybeans by M. A. Faldet. Similar extents of net protein escape were obtained with steam treatment at 100 or 110°C for about 30 min. The net protein escapes of hay protein treated with steam declined more rapidly than that of oven heated hay because of a more rapid increase in ADIN content. Both oven and steam heating of hays for the longer time periods increased ADIN to unacceptably high levels.

Estimated net protein escape in shredded hay was greater than that in normal hay ($P < .01$) when both hays were oven heated at the same temperature for same lengths of time. The reason for this is not known, but it is possible that the more rapid drying of shredded versus conventional hay resulted in preservation of greater amounts of soluble sugars, thus enhancing the Maillard reaction between sugars and protein during heating. Following steam treatment, ADIN in shredded hay was greater than normal hay, although net protein escape of the two kinds of hay was not different ($P > .05$).

Figure 1. Estimated rumen escape of protein in oven heated hay.

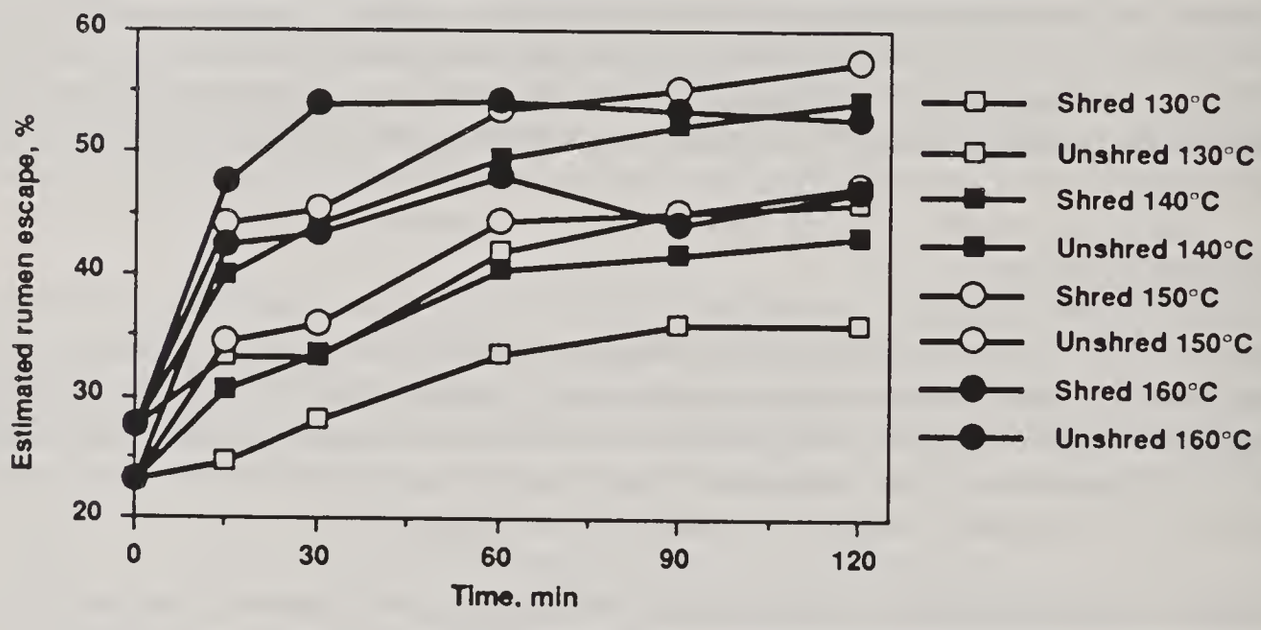
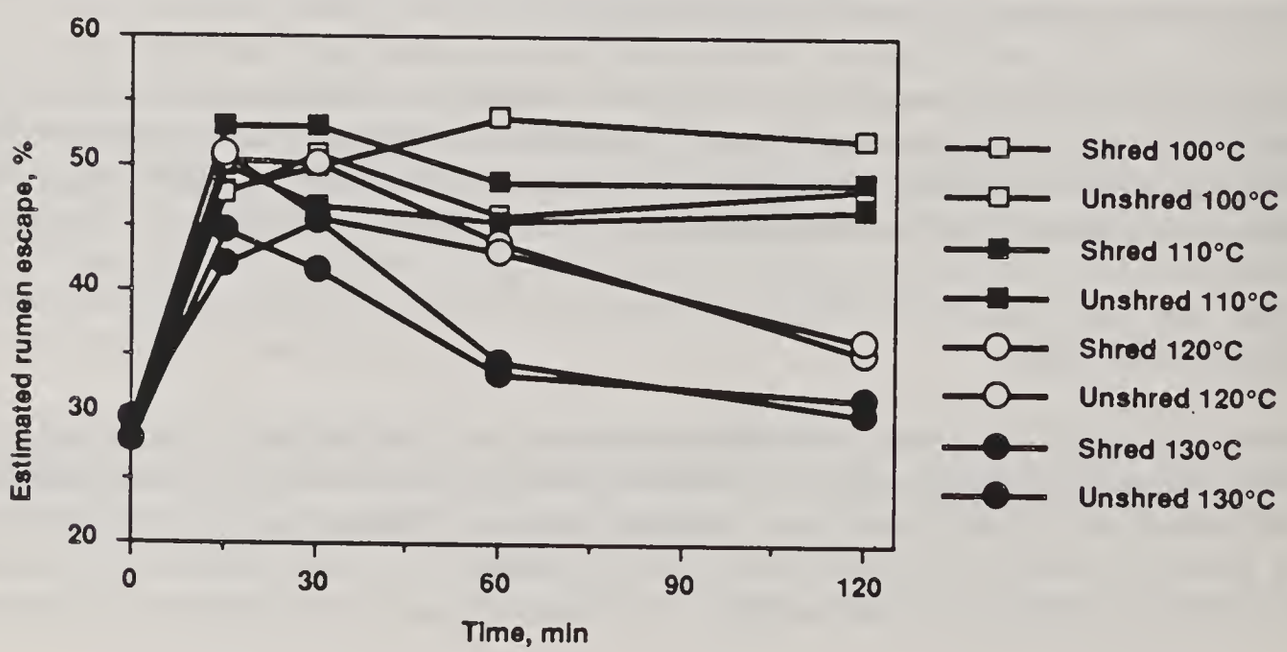


Figure 2. Estimated rumen escape of protein in steam heated hay.



CHEMICAL TREATMENTS TO IMPROVE ALFALFA SILAGE PROTEIN UTILIZATION

S.A. NAGEL and G.A. BRODERICK

Introduction

Alfalfa silage is valued for its high crude protein content; however, this protein is 75% or more degradable in the rumen. Chemical treatments of alfalfa silage may enhance its nutritional value by reducing protein degradation during ensiling and/or the rumen without greatly altering digestibility in the intestine. The purpose of this study was to determine whether formic acid or formaldehyde treatment of alfalfa silage would increase ruminal protein escape as indicated by 1) improved milk production and 2) altered milk production response to fishmeal supplementation.

Materials and Methods

Third cutting alfalfa was wilted to about 38% DM and three treatments were ensiled in polyethylene bags: control (C, no additive); formic acid additive (F, 90% w/v formic acid), applied at 8.2 l/ton; and Grainmax™ additive (G, 16% w/v formaldehyde), applied at 6.3 l/ton. Twenty-two Holstein cows received a 50% concentrate:50% alfalfa silage ration during a covariate period from days 4-18 after parturition. On day 19, cows were randomly assigned to a diet based on one of the experimental silages (C, F, or G). Rations were (DM basis) 98.48% silage, 0.7% dicalcium phosphate, 0.7% trace mineral salt, 0.12% Ketoban™, plus vitamins A, D, E. Cows were offered this ration ad libitum for six weeks, and data on milk production, milk component production, DM intake, and body weight change from the last five weeks were analyzed using the GLM of SAS. At week seven, the cows began a 4-week switchback experiment. Treatment groups were divided in half. Half received the same silage-based ration, the other half the same ration with fishmeal replacing 4.75% of the silage dry matter. Groups switched diets after two weeks. Milk production, milk component production, and DM intake data from the last week in each period were analyzed using the GLM of SAS.

Results and Discussion

Silage pH, ammonia and free amino acid (FAA) values are presented in Table 1. Compared to C, treatment F reduced the pH of the wilted material, and was most effective at preventing protein degradation in the silo, as shown by the low ammonia and FAA in the fermented material. Treatment G had lower ammonia and FAA values than C but values were significantly higher than F.

Dry matter intake was not different between the three groups (Table 2). Milk production was significantly increased in the F and G cows versus the C cows. The yield of solids-corrected milk also was lowest for C. Fat yield was highest in F and G groups, while protein yield was highest for F, intermediate for G, and lowest for C. Body weight changes were similar across treatments.

The switchback trial analysis compared fishmeal (FM) versus no fishmeal (No FM) within a silage treatment (Table 3). A response to rumen escape protein (FM) would indicate that the unsupplemented diet was protein deficient. There were no changes in DMI when FM was fed. Milk production and solids-corrected milk tended to increase with FM, but this was significant only for G.

Fat percent and yield were unaffected by FM addition. Protein percent and yield increased significantly for both C and G cows.

In summary, formic acid treated silage had lower pH, free amino acids, and ammonia when compared to either control or Grainmax treatments. Treatment of silage with either formic acid or Grainmax gave comparable improvements in milk production. Supplemental rumen escape protein (FM) tended to increase milk secretion; however, only Grainmax had significant increases. Milk protein secretion significantly increased in both control and Grainmax treatments when fishmeal was added.

Table 1. Effect of chemical treatment on silage pH, ammonia, and free amino acids.

	Treatment		
Wilted Material	C	F	G
pH	5.98[a]	4.27[c]	5.60[b]
Ammonia, % TN	0.37	0.21	0.45
Free amino acids, % TN	6.85	6.84	6.61
Fermented Material			
pH	4.49[a]	4.27[b]	4.43[a]
Ammonia, % TN	6.05[a]	1.32[c]	4.27[b]
Free amino acids, % TN	47.40[a]	24.13[c]	40.23[b]

[a,b,c]Means within rows with different superscripts differ ($p<0.01$).
C=control; F= formic acid; G=Grainmax (formaldehyde).
TN=Total nitrogen.

Table 2. Effect of diet on DMI, milk and milk component production, and body weight change.

	Ration			
Item	C	F	G	p
DM intake, kg/d	18.3	18.2	19.7	0.603
Body weight change, kg/d	-0.19	-0.26	-0.52	0.690
Milk, kg/d[1]	29.2[b]	32.6[a]	32.5[a]	0.022
SCM, kg/d	27.3[b]	31.2[a]	31.0[a]	0.040
Fat, %	3.71	4.06	4.03	0.104
Fat, kg/d	1.10[b]	1.30[a]	1.30[a]	0.026
Protein, %	2.74	2.90	2.68	0.126
Protein, kg/d	0.81[b]	0.92[a]	0.87[a,b]	0.100

[1]Covariate adjusted.
[a,b,c]Means within rows with different superscripts differ ($p<0.05$).
C=control; F= formic acid; G=Grainmax (formaldehyde).

Table 3. Effect of fishmeal supplementation on DMI, and milk and milk component production.[1]

	Ration					
	C		F		G	
Item	No FM	FM	No FM	FM	No FM	FM
DM intake, kg/d	21.6	20.1	21.1	22.1	23.3	22.2
Milk, kg/d	28.5	29.7	29.8	31.0	29.9*	31.3*
SCM, kg/d	24.4	25.2	27.2	27.8	25.9**	27.6**
Fat, %	3.15	3.07	3.54	3.44	3.30	3.36
Fat, kg/d	0.90	0.91	1.05	1.06	0.98	1.04
Protein, %	2.75***	2.87***	2.90	2.92	2.75***	2.86***
Protein, kg/d	0.78**	0.85**	0.85	0.90	0.82***	0.90***

[1]Statistical comparisons are between No FM and FM rations within a silage.
C=control; F= formic acid; G=Grainmax (formaldehyde).
*($p<0.10$), **($p<0.05$), ***($p<0.01$)

SUPPLEMENTATION OF HIGH FORAGE DIETS WITH ENERGY OR PROTEIN

T.R. DHIMAN and L.D. SATTER

Introduction

Cows fed diets containing a high proportion of alfalfa silage during early lactation produce less milk and milk with lower protein content than cows fed forage:grain in a 50:50 ratio. The purpose of this study was to determine the relative benefit of protein or energy supplementation for milk yield and milk composition in early lactating cows fed diets based on alfalfa silage.

Materials and Methods

Twenty-seven primiparous Holstein cows in early lactation (30-70 days postpartum) were randomly assigned to three treatments. An adaptation period of two weeks was given during which the cows were fed a pretrial diet (diet P) containing forage:grain in a ratio of 48.2:50. During weeks 3-9 of the experiment, cows were given experimental diets 1, 2 and 3 (Table 1). Daily feed intake and milk yield were recorded. Milk samples were analyzed for crude protein, NPN, lactose and urea. Body weights were taken once weekly after the morning milking. Weekly blood samples were analyzed for glucose, urea and ß-hydroxybutyrate (BHB). The milk protein composition of each animal was analyzed using polyacrylamide gel electrophoresis. Dietary intake of rumen degradable protein (RDP) and undegradable protein (UDP) were calculated by measuring in vitro protein degradabilities in alfalfa silage, shelled corn and fishmeal samples. Intake of NEL was calculated using NRC (1988) values for each feed ingredient (Table 3). The dry matter, crude protein, NDF and ADF contents of the alfalfa silage were 49.9, 19.5, 42.9 and 36.2% (dry basis).

Results and Discussion

Fat corrected milk yield was increased by 14% when fishmeal replaced alfalfa silage in an otherwise all forage diet. Addition of energy further increased the milk yield (Table 2). Milk fat content was slightly higher in Treatment 1 with the all forage diet, but there were no significant treatment differences. Milk crude protein content was raised by 0.18 and .41 units in Treatments 2 and 3 compared to Treatment 1 (Table 2).

Dry matter intake was higher with Treatment 3, whereas Treatments 1 and 2 supported similar intakes. Milk NPN, blood urea and milk urea were higher with Treatment 2, probably because of inefficient utilization of protein in the absence of adequate energy. Body weight gain during the experimental period was highest in Treatment 3 followed by Treatment 2 and 1.

Treatment 3 supported higher concentrations of blood glucose, whereas Treatments 1 and 2 had similar levels (Table 2). Treatments had no effect on the relative amounts of milk proteins.

According to NRC (1988) recommendations, Treatment 1 was sufficient in protein but was deficient in energy (Table 3). The results of this study indicate that the all silage diet which otherwise would be considered energy deficient is deficient in both protein and energy. It appears that the protein in alfalfa is overvalued due to the high degradability of alfalfa protein by rumen microbes.

Table 1. Ingredient composition of diet.

Ingredient	Diet			
	P	1	2	3
	.....% Dry Basis.....			
Alfalfa silage	48.2	98.2	91.2	61.2
Dry shelled corn	34.3	—	—	30.0
Soybean meal	15.7	—	—	—
Fish meal	—	—	7.0	7.0
Dicalcium phosphate	1.1	1.1	1.1	1.1
Trace mineral salt	0.7	0.7	0.7	0.7
Vitamin supplementation	trace	trace	trace	trace

Table 2. Treatment effects on milk yield, milk composition,dry matter intake and body weight change.

Parameter	Treatment 1	2	3	
Milk yield, kg/d	18.8[a]	22.2[b]	25.8[c]	(P=.0001)
3.5% FCM, kg/d	19.7[a]	22.5[b]	26.7[c]	(P=.003)
Milk fat, %	3.78	3.65	3.67	
Milk crude protein, %	2.85[a]	3.04[b]	3.26[c]	(P=.0002)
Milk NPN, mg/100ml	32.5[a]	36.7[b]	34.3[a]	P=.03)
Milk protein yield, g/d	529[a]	658[b]	845[c]	(P=.0001)
Milk fat yield, g/d	711[a]	797[b]	950[c]	P=.04)
Dry matter intake, kg/d	17.2[a]	17.4[a]	19.2[b]	(P=.05)
Body weight change, kg (wk 9 - wk 2)	7.0	15.1	19.7	
Milk lactose, %	5.04	4.91	5.02	
Milk urea, mM	6.6[a]	8.5[b]	6.7[a]	(P=.0001)
Blood urea, mM	6.1[a]	7.9[b]	6.2[a]	(P=.0001)
Blood glucose, mg/100ml	61.7[a]	61.7[a]	63.9[b]	(P=.04)
Plasma BHB, mg/100 ml	10.3	10.2	9.3	

Table 3. Dietary energy and protein intake.

Treatment	Diet component	NEL intake Mcal/d	RDP intake kg/d	UDP intake kg/d	Total Protein intake kg/d
1	Silage	22.0	2.16	1.14	3.30
2	Silage	20.6	2.03	1.07	3.09
	Fishmeal	2.1	.40	.39	.79
	Total	22.7	2.43	1.46	3.88
3	Silage	15.3	1.50	.79	2.29
	Fishmeal	2.2	.44	.43	.87
	Shelled corn	10.6	.31	.25	.56
	Total	28.1	2.25	1.47	3.72

VERIFICATION IN THE RUMINANT OF METHODS FOR OPTIMIZING HEAT TREATMENT OF SOYBEANS

M.A. FALDET, L.D. SATTER and Y.S. SON

Introduction

The amount of heat (temperature and time) needed for optimum treatment of soybeans has been identified using 1) an in vitro rumen procedure to determine protein degradation by rumen microbes and 2) two methods (Dinitro-flouro-benzene and rat growth) for estimating lysine availability. The purpose of this study was to confirm in vivo what the in vitro and chemical/rat growth studies were suggesting as the optimum treatment.

Materials and Procedures

Trial 1. Eight Holstein heifers weighing 410 kg were fed total mixed rations containing 79.5% alfalfa silage, 20% soybeans and 0.7% vitamin and mineral mix on a dry matter basis. Eight different heat treated soybeans were used: raw soybeans (raw), soybeans held for 3 h at approximately 120°C immediately after being roasted in a Gem Roaster (R+H), extruded soybeans (Extr), and soybeans jet sploded (JS). The remaining 4 treatments were obtained by changing the transit setting of a California pellet mill roaster. The temperatures of soybeans exiting the roaster were 117, 126, 138 and 154°C. An 8 x 8 Latin square experiment with 5 day periods was used. Jugular blood was taken on the last 2 d of each period 5 h post-feeding. Blood plasma amino acids were determined. Soybean treatments were analyzed for ruminal undegraded intake protein (UIP) by an in vitro rumen system and nutritionally available lysine (TLMI) by a chemical method. The product of UIP and TLMI was used to estimate the amount of lysine which escapes the rumen and is available for intestinal absorption (post-ruminal available lysine, PRAL). The feeding of optimally heat treated soybeans should result in the highest concentration of plasma branched-chain amino acids (BCAA).

Trial 2. Forty-four Holstein heifers weighing 150 to 250 kg were randomly assigned to one of four diets. Diets consisted of 91.8% alfalfa silage, .7% vitamin and mineral mix and 7.5% of one of four soybean treatments on a dry basis. Soybean treatments included raw soybeans, soybeans held for .5 h immediately after being roasted in a Gem Roaster (exiting soybean temperature of 141°C), and soybeans with and without .5 h holding with an exiting soybean temperature of 146°C. The two treatments of roasted soybeans were held at approximately 120-125°C. Heifer weights were taken on 3 consecutive days at the beginning and end of the 12 wk experimental period and weekly during the experiment. Jugular blood was taken on the last 2 d of wk 10 3 h post-feeding for blood plasma amino acid determination. Soybean treatments were analyzed for fractional rates of ruminal protein degradation, estimated UIP, and TLMI as described in Trial 1.

Results and Discussion

Trial 1. Dry matter and CP intakes were similar across diets averaging 8.5 and 2.2 kg/d. The average DM, CP, ADF and NDF content of the alfalfa silage fed was 31.5, 22.9, 32.1 and 39.1%. Results for UIP, TLMI, PRAL and plasma amino acid concentrations are given in Table 1. The R+H and Extr treatments resulted in the highest estimate of PRAL. This should result in the highest BCAA concentration in heifers fed those treatments. The highest BCAA concentrations resulted in heifers fed the R+H treatment. However, this was not the case for the Extr treatment. The

discrepancy could be related to an overestimate of UIP determined by the in vitro system due to oil inhibition of microbial protein degradation, since during the extrusion process oil vesicles are broken.

Trial 2. Dry matter and CP intakes were similar across diets averaging 7.0 and 1.43 kg/d, respectively. The average DM and CP content of the alfalfa silage fed was 52.3 and 19.1%. Results for UIP, TLMI, PRAL, average daily gain (ADG), and plasma amino acid concentrations are given in Table 2. Estimated PRAL was higher for soybeans roasted and held versus roasted or raw soybeans. However, body weight gain for heifers was similar across diets averaging .90 kg/d for 12 wks. Plasma amino acid concentrations were also similar across treatments. A high variability among heifers in growth and blood plasma amino acid response was observed.

Conclusion

Overall, animal results lend support for the laboratory analysis in identifying optimal heat treatment.

Table 1. Laboratory and plasma amino acid results from Trial 1.

Item	Raw	R+H	Extr	JS	117°	126°	138°	154°
UIP, %	24	64	57	39	44	47	53	61
TLMI, g/100g	2.30	1.79	2.06	2.28	2.24	2.18	1.96	1.54
PRAL, g/kg	5.52	11.5	11.7	8.89	9.86	10.2	10.4	9.39
BCAA[1]	543de	658^{a}	560^{cde}	583^{bcd}	541$_{e}$	569^{bcde}	607^{b}	594^{bc}
Lysine[1]	110	111	103	108	109	103	110	97

^{abcde}Means in the same row with different superscripts differ P<.05.

[1]Amino acids, nmole/ml plasma.

Table 2. Laboratory, heifer growth and plasma amino acid results from Trial 2.

		Holding, .5h			
Item	Raw-SB	146°	141°	146°	SEM
UIP, %	25.6	50.7	64.8	61.5	
TLMI, g/100g	2.28	2.18	2.08	2.02	
PRAL, g/kg	5.84	11.1	13.5	12.4	
ADG, kg/d	.91	.90	.89	.92	.085
BCAA[1]	684	650	731	700	33.5
Essential[1]	1273	1197	1354	1292	53.5

[1]nm/ml plasma

EFFECT OF FEEDING HEAT TREATED FULL FAT SOYBEANS ON PRODUCTION RESPONSES OF COWS IN EARLY LACTATION

M.A. FALDET and L.D. SATTER

Introduction

The protein in full fat soybeans is easily degraded by rumen microbes leading to surplus ammonia production in the rumen. Heat treatment can be a safe and economical means of decreasing protein degradation in the rumen. This study was conducted to assess the production response of early lactating cows fed either solvent soybean meal, raw soybeans or optimally heated soybeans with alfalfa silage as the sole forage.

Materials and Procedures

Heat treated soybeans (HSB, treated to maximize available lysine supply to the intestine), raw soybeans (SB), or solvent soybeans (SBM) were fed as protein supplements to early lactating cows. The HSB were roasted in a Gem Roaster (temperature of soybeans exiting roaster was 146°C) and held at 120°C for 30 min in 208 l barrels covered with canvas. The SB and HSB were cracked into halves and quarters before storing. Forty-six lactating cows (multiparous) were assigned sequentially to one of three diets on day 15 of lactation. Days 8 to 14 of lactation were used for a covariate adjustment of DM and CP intake and milk production. Data from the first week following the covariate period were not included in statistical analysis. Each of three treatment diets contained 50% alfalfa silage and 50% concentrate on a dry basis. The concentrate consisted of ground shelled corn, 2% vitamin-mineral mix and one of three protein supplements (10% SBM or 13% SM or 13% HSB) on a dry basis. The vitamin and mineral mix contained calcium supplements to obtain a calcium level of 1% of diet DM. All diets were formulated to provide 19% CP and were fed as total mixed rations. The SBM diet was fed during weeks 1 and 2 for covariate adjustment. Milk production and feed intake were measured daily. Each week body weights were taken and milk samples were analyzed for fat and protein.

Results and Discussion

Intakes of DM, DM as a proportion of body weight, and CP were similar across protein supplements (Table 1). Dietary CP content averaged 17.5% across treatment diets on a dry basis. Results for estimated rumen undegradable intake protein (UIP), available lysine, and estimated post-ruminal available lysine (PRAL) for diets are given in Table 1. The HSB had a higher estimate of UIP and PRAL compared to SBM or SB diets. Yields of milk, 3.5% fat-corrected milk, and protein were significantly higher for cows fed HSB compared to SB or SBM (Table 2). Composition of milk fat was not altered by treatment. However, supplementation of HSB or SB resulted in a reduction of protein content in milk compared to SBM. The decrease in milk protein content has also been reported by other researchers when feeding whole oil seeds or a fat source to lactating dairy cows. Body weight change was significantly lower for cows fed SB compared to SBM or HSB.

Overall, the increased yields of cows fed optimally heated soybeans could be attributed to improved protein nutrition. It is important to emphasize that the roasted soybeans used were exposed to more extensive heat treatment than is typical for soybeans roasted for commercial use.

Table 1. Results for DM and CP intake and actual composition of diets.

Item	SBM	SB	HSB
Intake,			
Dry Matter, kg/d	23.4	22.3	23.6
Dry Matter, % BW	4.06	3.76	3.84
Crude Protein, kg/d	4.11	3.91	4.11
Diet, % Crude Protein	17.6	17.6	17.4
UIP, % of diet CP[1]	34	31	42
Available lysine,			
g/100g TMR	.65	.94	.88
PRAL, g/100g TMR	.29	.27	.35

[1]Ruminal UIP used (%): Alfalfa silage, 25; Shelled corn, 58; SBM, 33; SB, 25; HSB, 65.

Table 2. Production response and body weight change.

Item	SBM	Raw SB	Ht-SB
Yield, kg/d			
Milk	34.5[a]	34.2[a]	38.9[b]
FCM, 3.5%	33.4[a]	34.7[a]	38.0[b]
Protein	1.01[a]	1.01[a]	1.10[b]
Milk composition, %			
Fat	3.41	3.50	2.41
Protein	2.99[a]	2.89[ab]	2.85[b]
Body weight change,			
kg/d	.25[a]	.04[b]	.28[a]

[ab]Means in the same row different superscripts differ P<.05.

EFFECT OF FORAGE TYPE AND CONCENTRATE LEVEL ON PERFORMANCE OF LACTATING DAIRY COWS

W.P. WEISS and W.L. SHOCKEY

Introduction

High quality alfalfa is considered the primary source of hay crop forage for lactating dairy cows in the United States and is considered the standard by which other forages are compared. There are conditions, however, when alfalfa may not be the most agronomically advantageous forage to grow. For example, alfalfa does not grow well in poorly drained soils and economically it is usually not practical to establish alfalfa in short rotation situations.

Orchardgrass is a high quality, cool season grass that can produce abundant amounts of high quality forage, especially in states with cooler climates. Because of high concentrations of neutral detergent fiber, which has been associated with feed intake limitations, grass species are generally discounted as quality feeds for dairy cows. However, many grasses grow well in poorly drained soils where alfalfa or other legumes grow poorly, and because of lower costs of seeding, are good choices for short rotation situations.

High quality alfalfa and orchardgrass silages were compared under three levels of concentrate feeding to determine to what extent quality grasses should be discounted when fed to lactating dairy cows.

Materials and Methods

Second and third cutting alfalfa was harvested in late bud stage, wilted to about 50% DM and ensiled into a glass-lined steel silo (49% DM, 21.7% CP, 40.1% NDF, 33.4% ADF, 1.50 Mcal NE_l/kg). Primary growth of orchardgrass was harvested in the vegetative stage of maturity and regrowth harvested every 25-28 d thereafter by wilting to about 40% dry matter then ensiling into a glass-lined silo (38% DM, 21.9% CP, 52.5% NDF, 32.7% ADF, 1.46 Mcal NE_l/kg). Orchardgrass was fertilized with 75 kg N/ha 5 times during the growing season at about 1 mo intervals. Both forages were chopped with the same harvester into 2 to 3 cm lengths. Forages remained in the silo approximately 1 yr prior to feeding.

Thirty Holstein cows (120 d in milk at the start of the experiment) were fed the two forages with differing amounts of a corn grain based concentrate mix (5 cows/treatment). Actual forage to concentrate (F:C) ratios were 82:18, 64:36, and 44:56.

Cows were individually housed and fed ad libitum once daily. After cows were fed diets for at least 30 d, a 5 d total collection digestion trial was conducted. Each week during the experiment, milk was sampled (AM and PM) and analyzed for milk fat and protein using spectrophotometry. Cows were weighed and feed was sampled.

Results and Discussion

Orchardgrass and alfalfa had similar concentrations of CP, ADF, and NE_l. As would be expected, orchardgrass had higher concentrations of NDF and hemicellulose. Neither forage would be considered heat-damaged (< 6% ADIN as % total N).

Results of the feeding and digestibility experiment indicate that, in contrast to the implication of our objective, orchardgrass should not be discounted when compared to alfalfa of similar quality. No forage x concentrate interactions were found for any parameter. Between forages the only statistical difference was for dry matter intake and dry matter digestibility. Dry matter intake was higher for alfalfa compared to orchardgrass. Cows produced milk with slightly higher efficiency when consuming the orchardgrass. Dry matter digestibility was higher for orchardgrass compared to alfalfa and may explain the observed improvement in efficiency.

Increased concentrate improved total milk and protein yield in 40 and 60% concentrate diets compared to 20% concentrate. Cows consuming the 20% concentrate diet had a higher % of fat in the milk than cows fed 40 or 60% concentrate. Although these differences are explainable on the basis of increased amounts of concentrates in the diet, the fact that no further improvement in milk or protein yield occurred in the 60 vs the 40% concentrate diets attests to the high quality of the forages fed. Due to the higher fat test of animals consuming the 20% concentrate diet, no difference was observed in fat-corrected milk yield.

Conclusions

High quality orchardgrass silage was substituted for alfalfa silage of a similar quality with no loss in performance. No benefit was derived from feeding 60 vs 40% concentrate and little benefit was observed in feeding greater than 20% concentrate with high quality forages to cows in mid to late lactation.

Table 1. Performance of cows fed different forages with different amounts of concentrate.

	FORAGE		CONCENTRATE		
	OG	ALF	20	40	60
Milk, kg/d	24.8	26.6	22.4	27.0	27.7
4% FCM, kg/d	22.2	23.6	21.2	23.7	24.0
Milk Fat, %	3.37	3.28	3.84	3.17	3.16
Milk Fat, kg/d	.82	.87	.82	.86	.86
Milk Protein, %	3.05	3.03	3.00	3.00	3.13
Milk Protein, kg/d	.75	.81	.67	.81	.86
DMI, kg/d	20.3	23.1	19.8	22.2	23.1
DMD, %	67.0	63.0	64.6	63.7	66.5
Body Weight, kg	545	569	555	553	562

RUMINAL SOLUBILIZATION OF SELECTED MACROMINERALS FROM FORAGES AND DIETS

D.R. LEDOUX and F.A. MARTZ

Introduction

Data on true absorption of minerals from forages are limited due to the very high costs and unique problems associated with in vivo animal studies. The development of alternative methods to predict mineral absorption would be of considerable benefit to the dairy industry. Since mineral absorption is a function of two processes, the solubilization of minerals in the digestive tract and the fractional absorption of minerals solubilized, a measure of mineral solubility may be useful in predicting absorption. The objectives of this study were to: (1) compare mineral solubility from diets with that of their respective forages; (2) determine if solubility is a limiting factor in the absorption of these minerals.

Materials and Methods

Feedstuffs evaluated and their nutrient composition are presented in Table 1. Feedstuffs were dried at 55°C and ground through a 1-mm stainless steel screen before being weighed into nylon bags. After a 10 d adaptation to diet (alfalfa hay, 15%; brome hay, 35%; corn silage, 50%), duplicate nylon bags (5 x 10 cm, 53+10 micron pores) containing feedstuffs (2 g) were placed into the rumens of two mature Guernsey cows for 0, 3, 12, 24 or 48 h. After retrieval, bags were washed, dried, weighed, and residues analyzed for Ca, Mg, K and P. Feedstuffs were analyzed for NDF and ADF using the Van Soest procedure. Disappearance rates and the proportion of minerals rapidly solubilized were determined by regressing the natural logarithm of mineral remaining in nylon bags on time. These values along with particulate passage rates were then used in the following formula to calculate effective ruminal solubility:

(1) $$\text{Effective Ruminal solubility} = A + (1-A)*K_d/(K_p + K_d),$$

where A is the proportion of minerals rapidly solubilized, K_d is the rate of mineral disappearance from nylon bags, and K_p is the particulate passage rate. The data were analyzed as a randomized complete block design with animals as a complete block and treatments arranged factorially (15 feedstuffs x 5 times).

Results amd Discussion

Forage and diet composition are presented in Table 1. Table 2 shows the proportions of DM and minerals that were solubilized in the rumen. Ruminal solubilization of DM averaged 50% for both forages and diets. Calcium solubility from forages was 12 percentage units greater (63%) than that from diets (51%). Since the greatest proportion of dietary Ca was supplied by the forage component of the diet this difference suggests that other dietary ingredients may have had a negative associative effect on total Ca solubility.

In contrast to Ca, there was a greater proportion (95%) of soluble K in diets compared to forages (91%) although the difference was considerably smaller. Potassium solubility values in excess of 90% for forages and diets are consistent with reports that this element is primarily associated with

the cell soluble fraction and occurs as the free ion. There were no differences between forages and diets with respect to P and Mg solubility which averaged 70 and 85%, respectively.

Solubility values for Ca and P from diets are compared to in vivo true absorption values in Table 3. With the exception of the FES-EL and A2-CS diets where in situ and in vivo values were similar, in vivo absorption values for Ca were 24 percentage units lower than in situ values (30 vs 54%). Particulate passage rates determined in this study averaged 5% h^{-1}, a rate lower than would be expected for a high producing dairy cow. However, even if passage rates were doubled, ruminal solubility values would only decrease by 10%. This suggests that Ca solubility does not limit absorption, especially since more Ca would be expected to be solubilized in the acidic portions of the lower tract.

Unlike Ca, there were four diets (FES-DC, CS-A, CS-B and A2-CS) where in vivo absorption values for P were higher than in situ solubility. However, in examining these diets we found that corn silage accounted for more than 30% of these diets and, coupled with previous reports indicating considerable P contamination of silage residues in nylon bags at 48 h, suggests that P solubility may have been underestimated. Results of this study indicate that solubility is not a limiting factor in the ruminant's ability to absorb these macrominerals.

Table 1. Composition of forages and diets.[1]

	NDF	ADF	Ca	P	Mg	K
	(%)					
Forages						
A1	56.1	37.9	1.44	.23	.17	2.92
A2	47.7	31.7	1.32	.30	.20	2.80
FES	59.3	27.5	.52	.31	.21	2.94
BR	62.2	36.4	.53	.11	.11	1.41
CS	40.3	18.3	.27	.22	.17	.74
Diets						
A1-DC	58.2	38.3	.88	.41	.17	1.83
A1-EL	45.9	21.6	.57	.38	.29	1.84
FES-DC	61.1	31.2	.47	.27	.19	1.92
FES-EL	47.1	22.0	.54	.46	.30	1.79
CS-A	50.3	25.8	.31	.18	.15	.77
CS-B	56.3	23.3	.18	.12	.10	.63
A2	45.8	26.5	.47	.17	.10	1.27
A2-CS	42.4	25.1	.35	.20	.13	1.04
BR	30.8	19.2	.23	.10	.06	.61
BR-A2	36.3	23.1	.36	.18	.11	1.04

[1]Dry matter basis. Forages are alfalfa (2 samples; A1,A2), fescue (FES), brome (BR) and corn silage (CS). Abbreviation for diets indicate their constituent forage(s).

Table 2. Effective ruminal solubilization of minerals.[1]

	DM	Ca	P	MG	K
			(%)		
Forages					
A1	46.2	54.8	55.2	80.9	91.1
A2	53.5	59.5	66.6	86.4	93.7
FES	53.6	70.7	78.4	88.0	93.2
BR	45.4	71.1	75.3	83.6	89.6
CS	54.5	59.4	81.6	91.2	86.7
Diets					
A1-DC	48.6	51.5	77.8	84.1	96.5
A1-EL	53.9	44.1	67.3	84.1	95.9
FES-DC	53.1	61.9	77.5	88.9	97.0
FES-EL	53.4	38.6	68.8	85.5	95.6
CS-A	51.8	59.1	76.0	90.3	93.9
CS-B	44.4	51.1	79.6	86.0	92.6
A2	47.1	58.0	64.4	84.0	95.8
A2-CS	53.8	45.3	71.6	87.2	95.6
BR	47.5	55.5	67.9	75.5	92.8
BR-A2	50.3	49.0	47.1	84.2	95.1
SEM	.7	1.1	2.1	.5	.2
LSD (P<.05)	2.2	3.4	6.5	1.6	.6

[1]Average passage rate and disappearance rates used in calculations were 5%/h for passage rate, and 2.6, 1.8, 1.3, 2.6, and 3.1%/h, for DM, Ca, P, Mg and K, respectively.

Table 3. Mineral solubility compared to in vivo true absorption.

	Ca		P	
	(%)		(%)	
	In Situ	In Vivo	In Situ	In Vivo
Diets				
A1-DC	51.5	25.4	77.8	55.6
A1-EL	44.1	28.1	67.3	62.8
FES-DC	61.9	42.9	77.5	94.4
FES-EL	38.6	37.0	68.8	66.1
CS-A	59.1	34.6	76.0	84.5
CS-B	51.1	43.8	79.6	93.9
A2	58.0	24.0	64.4	64.2
A2-CS	45.3	45.4	71.6	74.5
BR	55.5	20.0	67.9	46.0
BR-A2	49.0	17.0	67.1	63.0

EFFECTS OF SAMPLE SIZE AND SOLVENT ON IN VITRO CALCIUM SOLUBILITY

D.R. LEDOUX, A. GARCIA ORTIZ and F.A. MARTZ

Introduction

Data on mineral availability from forages are limited due to the very high costs and unique problems associated with in vivo studies. The development of an in vitro procedure to predict mineral availability from forages would be of considerable benefit to the dairy industry. The objectives of this study were to: (1) determine the extent of calcium solubility from five substrates; (2) determine if saturation is a factor when calcium solubility is measured by these procedures.

Materials and Methods

A completely randomized design with a 5x5x4 factorial arrangement of treatments was used. Substrates evaluated were alfalfa (A), corn silage (CS), and 3 mixed diets prepared using a commercial semipurified rat diet, solka floc, and three inorganic salts of Ca (calcium carbonate, diet 1; calcium oxalate, diet 2; dicalcium phosphate, diet 3). Solvents included: (1) 2X distilled water (W), (2) .01 N HCl (H), (3) .172% pepsin in .01 N HCl (HP), (4) rumen innoculum and McDougall's buffer (R), and (5) R followed by 3 h in pepsin-HCl (RP). For solvents 1-3, 50 ml of solvent (preheated to 39°C) were added to previously tared in vitro bottles containing the various sample sizes (.4, .8, 1.2 and 1.6 g) of ground substrates (1 mm screen) and the bottles capped, and placed into a water bath at 39°C for 3 h. For solvents R and RP, 50 ml of rumen innoculum and buffer solution (1:4 ratio and preheated to 39°C) were added to each in vitro bottle containing substrates and the bottles flushed with carbon dioxide, capped and placed into a water bath at 39°C for 48 H. For solvent RP, at the end of 48 h, 6 ml of HCl and 2 ml of pepsin (5%) were added to each bottle which was then returned to the water bath for an additional 3 h. Calcium solubility was determined from the ratio of mineral in total digest supernatant (samples centrifuged at 18000x g for 15 min) to that supplied by the substrate. Data were analyzed by analysis of variance by the GLM procedures of SAS.

Results and Discussion

Effects of sample size and solvent on Ca solubility from substrates are shown in Table 1. Across both solvent and sample size, Ca from CS was most soluble (83%) followed by Ca from diet 3 (73%), diet 1 (68%), A (45%), and diet 2 (44%). With the exception of A, where the lowest Ca solubility was observed for solvent R, Ca was the least soluble in solvent W. Overall, Ca was the most soluble in solvent RP, the exception being diet 2 where Ca was most soluble in solvent R. Calcium solubility values determined in this study suggest that solubility is not a limiting factor in the ruminant's ability to absorb Ca since solubility values for solvent RP (a solvent that simulates in vivo mineral digestion) all exceed the in vivo true absorption value (38%) currently used by NRC (1988). In order to determine if saturation is a factor in Ca solubility, soluble Ca was plotted against added Ca for all substrates. A decrease in percent soluble Ca with increasing amounts of added Ca is indicative of saturation. With the exception of diet 1 and 3, where Ca solubility in solvent H began to approach saturation at a sample size of 1.2 g, saturation did not appear to be a factor in determining Ca solubility by these methods.

Table 1. Effect of sample size and solvent on Ca solubility (%).

	SOLVENTS				
S SIZE(g)	W	H	HP	R	RP
		ALFALFA[a]			
0.4	33.4	67.6	64.4	0.0	83.9
0.8	35.5	57.5	54.1	0.0	82.8
1.2	36.7	49.1	48.6	0.4	85.6
1.6	38.1	45.0	47.5	0.2	82.9
L[b]	NS	**	**	NS	NS
Q[c]	NS	NS	NS	NS	NS
		CORN SILAGE[a]			
0.4	60.6	84.9	75.9	85.4	100.0
0.8	67.4	87.1	81.7	67.7	91.7
1.2	67.5	84.2	81.0	100.0	99.9
1.6	64.2	77.7	77.1	100.0	99.7
L[b]	NS	**	NS	**	NS
Q[c]	**	NS	NS	**	NS
		DIET 1[a]			
0.4	12.0	93.3	93.2	10.1	93.6
0.8	11.8	100.0	97.0	57.5	100.0
1.2	8.1	91.5	83.2	75.4	100.0
1.6	7.6	70.8	80.2	76.5	100.0
L[b]	NS	**	**	**	**
Q[c]	NS	**	NS	**	**
		DIET 2[a]			
0.4	0.0	19.5	16.3	97.9	65.2
0.8	0.0	38.6	25.5	100.0	60.8
1.2	0.0	42.6	24.2	100.0	59.8
1.6	0.0	39.2	22.3	100.0	51.4
L[b]	NS	**	NS	NS	**
Q[c]	NS	**	**	NS	NS
		DIET 3[a]			
0.4	0.0	81.6	74.1	83.4	96.9
0.8	6.0	95.3	83.9	97.3	94.8
1.2	8.2	93.5	77.1	100.0	100.0
1.6	9.7	80.1	69.0	100.0	100.0
L[b]	**	NS	NS	**	NS
Q[c]	NS	**	**	**	NS

[a]LSD (P<.O5) = 7.1 for any two interaction means.
[b]Linear orthogonal contrast.
[c]Quadratic orthogonal contrast.

TRUE ABSORPTION OF CALCIUM AND PHOSPHORUS FROM ALFALFA HAY, BROMEGRASS HAY AND CORN SILAGE FED TO LACTATING DAIRY COWS

F.A. MARTZ, A.T. BELO, M.F. WEISS and D.R. LEDOUX

Introduction

Little information exists about true absorption of calcium (Ca) and phosphorus (P) from alfalfa, bromegrass and corn silage when fed to lactating dairy cows. The objectives of this study were: (1) determine the true absorption of (Ca) and (P) from these forages when fed to lactating dairy cows (2) determine true absorption of calcium and phosphorus when fed at levels below nutritional requirements.

Materials and Methods

The experimental design was a crossover with two dietary treatments, alfalfa-bromegrass hay (AB) and bromegrass hay (B), four cows with repeated measures on cows. At the end of period 2 all cows were fed a CS ration for a 3-week period.

Each period consisted of a 14-day adaptation period and a 7-day sample collection period. Feed intake, feces, urine, and milk output were measured during the collection period. Each cow was dosed intravenously with Ca^{45} and P^{32} on day 1 of each period. Blood was sampled at varying time intervals during the subsequent 6 days.

Four cows, in lactations 6 and 7, which were producing 38 to 42 kg of milk daily were randomly assigned to treatments. Cows had been lactating 60 to 90 days and were fed CS, alfalfa hay, soy hulls, whole cotton seeds and concentrate prior to this trial. Their Ca and P intake were calculated to be 150 and 75 g/d, respectively.

Diets (Table 1) were equal in energy, protein, vitamins and minerals except Ca and P. Rations were also formulated so that a major portion of the Ca and P came from alfalfa, bromegrass or corn silage. Hominy grits and corn gluten meal were used as sources of energy and protein and were relatively free of Ca and P. Differences among means were determined by paired T tests.

Results and Discussion

Average BW of cows was 634 kg. Intake of DM was similar for rations AB and B (21.6 & 20.1 kg/d) but CS was less ($P<.05$) (19.9 kg/d). Apparent digestibility of DM was equal for rations AB, B, and CS (66.9, 64.1, 66.1 %). Fat corrected milk was similar for AB and B (36.6 & 33.4 kg/d) but less for CS ($P<.07$) (33.0 kg/d). Daily intake of Ca and P were very low for high producing dairy cows (Table 2); 50% or less of the current NRC requirement for all diets. Since we used these same sources of alfalfa and corn silage in previous trials, true absorption values determined in this study can be compared with previous determinations. We were surprised at the low true absorption values for Ca in rations AB (17.2%) and B (20.4%). Previous partial true absorption values for Ca in alfalfa of AB was 24% which is similar to the value for this trial. Data also indicate the true absorption for Ca from bromegrass was near 20%. True absorption of Ca in CS (46%) is similar to previous determinations (51%).

Plasma 1,25 dihydroxy Vitamin D is indicative of a cow's ability to absorb Ca. Plasma 1,25 D was higher (P<.02) for CS than AB or B (Table 2) and AB and B are similar to values observed in a previous trial where alfalfa and CS were compared and true absorption was similar to this trial.

True absorption of P in this trial was lower for AB (53.4%) and CS (65.7%) than previous trials for alfalfa (64%) and corn silage (74% and 85%).

Recent reports indicate that rations containing fixed cation/anion balance of over 50 meq/100g DM were disruptive to Ca absorption and metabolism. Rations in our trial had a calculated balance of near 50 meq/100g DM. Differences in true absorption of Ca did not appear to relate to fixed cation/anion balance of rations.

Table 1. Formulation of alfalfa-bromegrass hay (AB), bromegrass hay (B) and corn silage (CS) rations fed to lactating cows (DM basis).

	Percent DM			Ca Supplied (g/d)			P supplied (g/d)		
Ingredient	AB	B	CS	AB	B	CS	AB	B	CS
Brome	24.1	43.4	—	20.0	37.0	—	5.0	9.0	—
Alfalfa	24.1	—	—	66.0	—	—	17.0	—	—
C. silage	—	—	60.2	—	—	34.0	—	—	25.0
Hominy	44.6	45.4	29.0	1.0	1.0	—	10.0	10.0	6.0
C.G. Meal	5.8	9.5	8.9	1.0	2.0	2.0	6.0	10.0	9.0
Urea	.8	1.1	1.0		—	—	—	—	—
Salt, TM[1]	.4	.4	.4		—	—	—	—	—
Dynamate[2]	.2	.2	.2		—	—	—	—	—
Bicarb	—	—	.3		—	—	—	—	—
Total	100.0	100.0	100.0	88.0	40.0	36.0	37.0	29.0	41.0
Fixed C/A[3]	57.6	51.7	54.1						
Fixed C/A[4]	40.2	39.8	42.8						

[1] Trace mineral salt: Mg 2,000; Fe 5,000; Zn 2,000 ppm.
[2] Dynamate: Trademark by IMC, Inc., Mundilene, Illinois. K 18, Mg 11, S 22%.
[3] Calculated fixed cation/anion balance [(Na + K)-(Cl)] meq/100 g DM.
[4] Calculated fixed cation/anion balance [(Na + K)-(Cl + S)] meq/100 g DM.

Table 2. Calcium and phosphorus balance, true absorption and plasma 1,25 Vitamin D for cows consuming alfalfa-bromegrass hay (AB), bromegrass (B) and corn silage (CS) rations.

Measure	Ration			Pooled	P
	AB	B	CS	SEM	Value
Calcium					
NRC req., g/d	146.0	146.0	146.0	—	—
Intake, g/d	78.2[a]	37.0[b]	36.7[b]	4.9	.01
Fecal, g/d	77.6[a]	42.6[b]	25.9[c]	5.8	.01
Urine, g/d	4.5[a]	12.3[b]	4.9[a]	2.2	.01
Endogenous, g/d	12.8[a]	13.3[a]	6.3[b]	1.8	.02
Milk, g/d	23.7[a]	23.3[a]	31.1[b]	2.4	.01
Balance, g/d	-27.6[a]	-41.3[b]	-25.1[a]	5.3	.02
Appt. absorption, g/d	- 0.6[a]	5.7[a]	10.8[b]	4.3	.02
True absorption, %	17.2[a]	20.4[a]	46.4[b]	9.9	.01
Pls. 1,25 vit D, pg/ml	58.1[a]	68.1[a]	103.9[b]	12.6	.02
Pls. hyd. proline, mg/ml	2.2	2.7	2.5	.3	NS
Phosphorus					
NRC reg., g/d	81.0	81.0	81.0	—	—
Intake, g/d	41.8[a]	28.8[b]	44.3[a]	3.4	.01
Fecal, g/d	24.0[a]	21.9[ab]	20.0[b]	2.7	.01
Urine, g/d	.1	.1	.1	.02	NS
Endogenous, g/d	4.4	6.4	4.9	2.2	NS
Milk, g/d	22.7	21.5	23.7	1.8	NS
Balance, g/	- 4.9[a]	-14.8[b]	.5[c]	2.8	.05
Appt. absorption, g/d	17.9[a]	6.8[b]	24.3	2.5	.1
True absorption, %	53.4[a]	46.0[b]	65.7[c]	6.3	.05

[abc] Means in same row with different superscripts differ.

MILK PRODUCTION IN EARLY LACTATION FROM FIRST CUTTING ALFALFA CUT AT TWO MATURITIES AND HARVESTED AS SILAGE OR HAY

W.F. NELSON and L.D. SATTER

Introduction

This experiment is the fifth in a series of trials, each utilizing forty or more lactating cows, that we have conducted to quantify the effects of alfalfa maturity and method of preservation on milk production. Results from previous studies indicate that mid lactation cows producing less than 25 kg/d of milk are not adversely affected by increasing alfalfa maturity from early bud to early flower. When early lactation cows averaging up to 35 kg/d of milk were used, the impact of maturity was observed to be a decrease of 0 to .15 kg milk/cow/day for each day of delayed alfalfa harvest beyond early bud stage. Cows receiving alfalfa as silage in 60:40 forage:grain ratio produced 1.5 to 2.5 kg/d more milk than cows fed 60:40 alfalfa hay - based diets.

Materials and Methods

Two maturities of first cutting alfalfa were harvested in 1988. Half of each maturity was preserved as silage in horizontal concrete bunker silos, and half was preserved as dry hay in small square bales. Table 1 contains morphological and chemical profiles of the four forages harvested. Forty eight cows (8 primiparous and 40 multiparous) were assigned to one of the four forage treatments on day 15 of lactation subsequent to a two week covariate period. The covariate diet was an alfalfa silage - based ration containing 19% CP and 30% NDF. The treatment period lasted 10 weeks. Each of the treatment diets contained 60% alfalfa [early cut silage (ECS), early cut hay (ECH), late cut silage (LCS) or late cut hay (LCH)] and 40% grain mix on a dry basis. Silage diets were fed as total mixed rations once daily. Cows on hay diets received hay once daily in hanging hay feeders placed in the manger, while grain was fed twice daily. All diets were formulated to provide 19% CP by adjusting the proportions of corn and soybean meal. Milk production and feed intake were measured daily. Body weights were taken and milk samples were analyzed for fat and protein weekly.

Results and Conclusions

Data from week two of lactation were used for covariate adjustment of milk production. Milk production values from weeks 4 through 12 of lactation were adjusted for the covariate period and are shown along with actual mean milk production values in Table 2. Intake and milk composition parameters were not adjusted for the covariate period. Increasing maturity of alfalfa had little effect on milk production of cows yielding in excess of 35 kg/d. The greatest effect of maturity observed was a 1.4 kg/d difference between the ECS and LCS diets (decrease of .10 kg milk/cow/day per day of delayed harvest). This difference is not statistically significant. No decrease in milk production was observed with increasing alfalfa maturity for the hay diets.

When milk production values were adjusted for the covariate period, cows consuming silage diets produced significantly more milk than cows fed hay diets (2.8 kg/d more milk). The ability of cows on silage diets to outperform cows on hay diets in this experiment agrees with results from two earlier trials. Since no differences in dry matter intake were observed between silage and hay diets in

this experiment, the data suggest that the dry matter in diets containing alfalfa silage was more efficiently utilized than the dry matter in diets containing alfalfa hay of similar quality. The method of feeding (total mixed rations for silage diets; hay and grain separately) may be an explanation for the milk production differences between alfalfa silage and alfalfa hay. We are continuing to study the efficiency of dry matter and fiber utilization in diets containing alfalfa silage and alfalfa hay.

Table 1. Forage maturity and composition.

	Date cut	Mean stage by weight	%CP	%NDF	%ADF	%ADL
Early cut silage	5/24/88	2.3	21.8	38.5	32.8	6.3
Late cut silage	6/06/88	4.3	17.3	45.3	38.1	7.2
Early cut hay	5/24/88	2.3	20.0	39.7	33.6	6.4
Late cut hay	6/06/88	4.3	15.2	46.2	40.8	7.8

Table 2. Response of early lactation cows to four alfalfa forages fed as 60% of the ration dry matter. (Values are means of observations from week 4 through 12 of lactation.)

	Actual Milk kg/d	Adjusted Milk kg/d	Milk Fat %	Milk Prot %	DMI kg/d	DMI % of BW	NDFI kg/d	NDFI % of BW
ECS	38.2	38.1	3.64	2.78	21.7	3.68	5.5	.94
LCS	36.8	37.0	3.45	2.82	22.6	3.91	6.7	1.16
ECH	34.3	35.0	3.53	2.90	22.6	4.11	5.9	1.07
LCH	35.9	35.0	3.55	2.80	21.9	3.87	6.7	1.18
p values,								
EC vs LC	.95	.53	.27	.58	.90	.97	.01	.01
S vs H	.17	.01	.92	.31	.91	.16	.60	.08

IMPACT OF STAGE OF MATURITY AND METHOD OF PRESERVATION OF ALFALFA ON FACTORS AFFECTING DIGESTION AND PASSAGE IN LACTATING DAIRY COWS

W.F. NELSON and L.D. SATTER

Introduction

A series of experiments has been conducted to measure the impact of alfalfa maturity and preservation as silage or hay on milk production of lactating cows fed isonitrogenous 60:40 forage:grain rations. Depression in milk production due to maturity has been modest. However, cows have consistently produced more milk when fed total mixed rations containing alfalfa silage than when hay of similar quality is fed separately from grain. The purpose of this experiment was to measure the impact of maturity and preservation of alfalfa on digestion in the ruminant GI tract.

Materials and Methods

Four multiparous Holstein cows fitted with rumen cannulae and in mid lactation were fed 60:40 forage:grain (dry basis) rations containing first cutting alfalfa (1988) harvested at the early bud (5/23) or early flower (6/6) stage of maturity. Each alfalfa maturity was preserved as silage and hay. All cows were fed isonitrogenous 60:40 forage:grain rations. Grain was fed four times daily. A 4x4 Latin Square design (28 d periods) was conducted to measure the impact of alfalfa maturity and method of preservation on milk production, intake, chewing activity, rumen fill, digestibility, rumen retention time, rate of alfalfa digestion and particle size of digesta. Rumens were manually emptied, dacron bags were used to measure forage rate of digestion and markers were used to measure rumen retention time. A 14 d slow-release product (Sometribove, Monsanto) of recombinant bovine somatotropin was used to increase milk production and, thus, energy demand throughout the experiment.

Results and Conclusions

Milk production and composition were not significantly affected by treatment. Increases in fiber intake resulted due to increased maturity and preservation as hay, but did not appear to limit dry matter intake or milk production. Both increased maturity and preservation as hay resulted in more time spent ruminating, greater total chewing time, greater wet and dry rumen fill and a greater volume of rumen contents. Rumen retention times of Lanthanum (applied to forage) averaged 6 hours less and 0 h disappearance of dry matter from dacron bags was greater for silage than hay. In situ rates of dry matter disappearance averaged 15%/h for silage and 9.5%/h for hay. Lag time was inversely related to 0 h disappearance. Masticates and mixed rumen samples from cows fed hay showed a greater percentage of dry matter to be particles > 9.5 mm in length. Cows produced milk more efficiently when fed alfalfa silage compared to alfalfa hay due to more rapid digestion and passage. The potential for rumen fill to limit intake in high producing cows appears to be greater for alfalfa preserved as hay than for alfalfa preserved as silage.

Table 1. Chemical composition of early bud and late bud alfalfa silage and hay and composition of 60:40 diets (dry basis).

	DM(%)	CP(%)	NDF(%)	ADF(%)	ADL(%)	MSW[a]
Forages;						
Early cut silage	31.0	22.4	37.9	33.5	6.3	2.3
Late cut silage	32.0	17.4	45.1	39.0	7.6	4.3
Early cut hay	88.0	21.3	42.1	31.8	6.4	2.3
Late cut hay	89.8	15.5	47.8	39.2	7.5	4.3
Diets;						
Early cut silage	42.0	19.8	26.6	21.8	4.2	
Late cut silage	43.1	18.6	31.1	25.4	4.9	
Early cut hay	88.5	19.7	29.2	20.8	4.2	
Late cut hay	89.6	18.2	32.9	25.7	4.9	

[a] Mean stage by weight.

Table 2. Animal response in cows fed alfalfa silage or hay harvested at two stages of maturity.

	Diet			
Item	ECS	LCS	ECH	LCH
Milk, kg/d	30.1	27.5	30.0	30.5
Milk fat, %	3.90	3.78	3.60	3.83
Milk protein, %	3.03	3.25	3.09	3.09
4% FCM, kg/d	29.6	26.6	28.2	29.7
Dry matter digestibility, %	62.4	56.6	61.2	56.0
Intakes;				
DM, kg/d	20.7[b]	21.9[ab]	23.8[a]	23.3[ab]
DM, % of BW	3.2[b]	3.4[ab]	3.7[a]	3.5[a]
NDF,kg/d	5.2[b]	6.5[a]	6.6[a]	7.4[a]
CP, kg/d	4.14.1	4.6	4.4	
Chewing time;				
Ruminating, min/d	292[b]	392[a]	389[a]	469[a]
Total, min/d	682[b]	822[a]	817[a]	883[a]
Rumen fill;				
Wet, kg	79.6[c]	86.6[b]	88.8[ab]	91.0[a]
Dry, kg	9.9[c]	12.4[b]	13.8[a]	14.0[a]
Volume, l	97.8[c]	108.4[b]	114.1[ab]	118.1[a]
Rumen retention time of markers;				
Forage (La), h	18.9[ab]	17.5[b]	23.2[a]	25.7[a]
Grain (Sm), h	17.018.8	21.3	23.0	
Liquid (Cr), h	17.1	15.2	16.2	18.2
In situ parameters;				
0 h residue, %	46.8[a]	59.7[b]	64.5[b]	65.0[b]
Indigestible, %	18.8[b]	27.5[a]	20.8[b]	27.4[a]
K, %/h	16.0[a]	13.9[a]	8.1[b]	10.8[b]
Particles > 9.5mm, %				
Boluses	23.8[b]	26.7[ab]	25.7[ab]	30.9[a]
Rumen contents	12.0[ab]	8.9[b]	15.8[a]	14.3[a]

[abc]Numbers within row without a common superscript differ (P<.05).

EFFECT OF SPECIFIC GRAVITY OF ALFALFA HAY AND SILAGE ON RUMEN STRATIFICATION

M. A. WATTIAUX, L.D. SATTER and D. R. MERTENS

Introduction

The sorting and sifting of particles for passage from the reticulo-rumen is influenced by rumen motility and digesta characteristics. Usually there is a "solid mat" of large, light particles in the dorsal region of the rumen, and a ventral "liquid pool" rich in small, dense particles. The separation of digesta tends to isolate the pool of particles eligible for passage in the vicinity of the reticulo-omasal orifice. Previous in-vitro experiments showed that alfalfa silage (AS) particles are heavier than alfalfa hay (AH) particles during digestion. Heavier particles might be freed from the dorsal mat more rapidly than lighter particles. In this experiment, stratification of dry matter (DM) in the rumen was measured and related to the specific gravity (SG) of forage particles during in-vitro digestion.

Materials and Methods

Three mid lactation Holsteins cows fitted with rumen and duodenal cannula were used in a modified 4X4 Latin square. Cows were fed four times daily diets including forages (AH or AS: 79% DM basis), soybeans (heated or raw: 19%), and a mineral-vitamin mix (2%). After 14 days of adaptation, a 220 ml bottle was used to sample digesta from the ventral rumen and the mid reticulum. Grab samples were also taken from at least three locations in the dorsal region of the rumen. In each of the five periods, DM percentages were determined on 12 samples collected from each location over a period of 48 h and oven dried at 60°C. Four hundred ml of duodenal digesta were collected every 4 h for 72 h, and DM content was determined by freeze drying.

Results and Discussions

In agreement with in-vitro hydration studies, AH particles retained more water than AS particles in the dorsal region of the rumen (Table 1). With the hay based diets, digesta in the ventral rumen and reticulum was of lower DM content which might reflect more saliva secretion. However, duodenal digesta had a higher percentage of DM with the hay based diets. The 18 and 24% increase in concentration of DM in the ventral rumen and reticulum, and the lower ratio of DM in the dorsal/ventral rumen for the silage based diets suggest that the denser AS particles (Figure 1) sink faster into the ventral rumen. Assuming that the quantity of digesta DM flowing through the reticulo-omasal orifice per reticular contraction is proportional to the concentration of DM in the reticular contents, the results of this experiment indicate that more DM can flow per reticular contraction with AS than AH based diets.

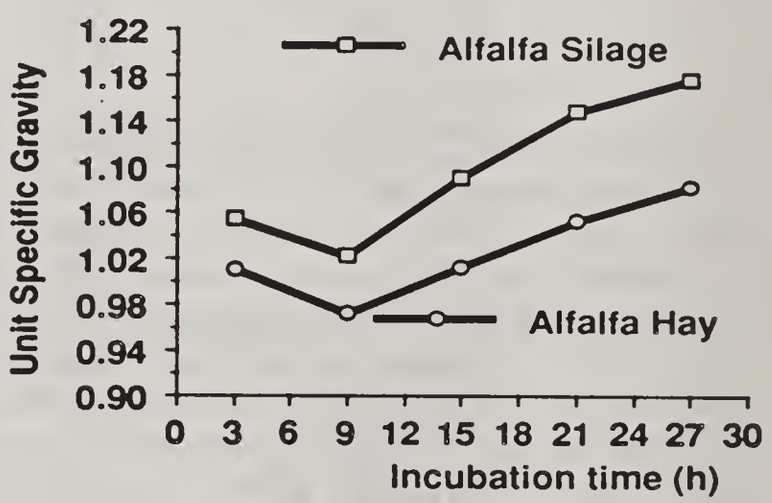


Figure 1. Change in unit specific gravity of alfalfa hay and alfalfa silage particles during in-vitro digestion.

Table 1. Percent DM in rumen and duodenal samples when cows are fed alfalfa hay or alfalfa silage based diets.

Items	Diets		Ratio	p level
	AH	AS	AS/AH	
Location				
Dorsal (g DM/100 g)	17.4	18.1	1.04	<.03
Ventral (g DM/100 ml)	5.3	6.3	1.18	<.03
Reticulum (g DM/100 ml)	4.9	6.0	1.24	<.002
Duodenum (g DM/ 100 g)	4.6	4.2	0.91	
Ratio				
Dorsal/Ventral	3.3	2.8		
Dorsal/Reticulum	3.6	3.0		
Ventral/Reticulum	1.1	1.0		

EFFECT OF DRY MATTER SOLUBILIZATION, WATER OF HYDRATION, AND GAS ENTRAPMENT DURING DIGESTION ON THE SPECIFIC GRAVITY OF GRASS AND LEGUME FORAGES PRESERVED AS HAY OR SILAGE

M. A. WATTIAUX, L. D. SATTER and D. R. MERTENS

Introduction

Entrapment of gases during fermentation lowers the functional specific gravity (FSG) and possibly delays the passage of partially digested particles. However, solubilisation of dry matter (DM) and water of hydration, which are not included in the FSG measurement, also affect specific gravity (SG) and hence the eligibility of particles for escape from the reticulo-rumen. A series of experiments were conducted to measure SG of particulate matter (DSG) and to assess the altering effects of water of hydration and gas entrapment during in-vitro digestion of alfalfa hay (AH), alfalfa silage (AS), and bromegrass hay (BH).

Materials and Methods

In the first experiment, samples of forages ground through a 2 mm screen were placed in pycnometers partially filled with degassed distilled water and DSG was measured after degassing under vacuum at 720 mm Hg for 30, 60, 90, and 120 min. The insoluble DM was recovered by centrifugation and oven drying. In the second experiment the total amount of water bound to the particles was measured, and this was the subject of an earlier report. The SG of the hydrated particles is referred to as Compact SG (CSG). In the third experiment, the FSG of digesting particles

was measured using pycnometers as incubation flasks. Results of these experiments were combined in an attempt to calculate the Unit SG (USG) which includes the effect of the solid, liquid, and gas fractions that make up a particle:

$$USG = DSG * S + 1 * W + 0 * G$$

Where: DSG is the SG of particulate DM
S is the fractional volume of residual particulate DM
W is the fractional volume of water of hydration
G is the fractional volume of gas during fermentation
with $S + W + G = 1$

Assumptions made in calculating USG are: 1) DSG does not change during digestion; 2) the SG of water of hydration is 1.00; 3) there is a 20 % reduction in the total bound water due to digestion (Mc Burney et al., 1985); and 4) the gas associated with particles during digestion does not substitute for water, but increases the functional volume of the particles. This is because the bubbles are thought to be nucleated at the outer surfaces of particles.

Results and Discussions

Thirty min. of vacuuming was sufficient to remove gases from the pore spaces of the particles ($P<.05$). After DM solubilisation occured, the SG of the water (1.00) in the pycnometer increased to 1.0050, 1.0044, 1.0021 for AS, AH, and BH, respectively. The higher the solubility of the DM, the lower the SG of the residual particulate DM, suggesting that the soluble DM has a higher SG than the fibrous fraction of the feed.

Per unit of volume, gases have a greater impact than water in altering the SG of the residue. However, USG depends on the proportion of the total volume occupied by water and gas associated with the particles. At 9 h of digestion, the fractional volume of gas and water were: .158, .589; .134, .543; .138, .507, for AH, AS, and BH, respectively. Despite a higher fractional volume of water, the gas alone is responsible for most of the decrease in SG of the residual DM, as FSG and USG are similar (Table 1). However, at 27 h of digestion, the fractional volume of gas and water were: .062, .656; .001, .616; .093, .533, respectively. At this point, water was the main factor altering the SG of the residual DM as CSG and USG are similar.

Conclusion

This series of experiments indicates that solubilisation of DM must be accounted for in measuring the DSG of forages. The effect of gas entrapment is an important factor during active fermentation, and might be responsible for a delayed outflow from the rumen for particles. However, USG at the end of the digestion period was determined primarily by the amount of water of hydration associated with the residual DM.

Table 1. Effect of water of hydration and entrapment of gas during digestion on SG of three forages

Items	Effect accounted for in SG calculation	AS	AH	BH
DSG	**DM (total)**	**1.60[a]**	**1.60[a]**	**1.56[b]**
Soluble DM (%)		34.4	26.8	13.0
DSG	**DM (particles)**	**1.47[a]**	**1.50b**	**1.52[c]**
Bound water (ml/g DM)		1.14	1.55	.94
CSG	**DM+Water**	**1.17**	**1.15**	**1.21**
Gas Volume (ml/gDM)				
9 h. of digestion		.28	.42	.25
27 h		.00	.15	.16
FSG 9 h	**DM+Gas**	**1.05[a]**	**.93[b]**	**1.10[c]**
27 h		**1.47[a]**	**1.23[b]**	**1.23[b]**
USG 9 h	**DM+Water+Gas**	**1.02**	**.97**	**1.05**
27 h		**1.17**	**1.08**	**1.10**

[a, b, c]Means in the same row differ ($p<.05$)

EFFECT OF FORAGE PRESERVATION METHOD AND PROTEIN SUPPLEMENTON OPTIMAL DIETARY NDF LEVELS

R.G. DADO, D.R. MERTENS, G.A. BRODERICK, and R.W. HINTZ

Introduction

Previous research has shown that optimal concentrations of neutral detergent fiber (NDF) in rations for lactating dairy cows result in energy densities and feed intake potentials that maximize milk production. These ration NDF optima vary according to stage of lactation and level of production. Fiber utilization depends on fermentation activities in the rumen. Because this activity and maximum milk production can be affected by amount and type of protein, it was hypothesized that optimal NDF levels also vary according to amount and degradability of dietary protein. The objective of this experiment was to determine if higher optimal levels of dietary NDF are obtained when protein degradability is altered by varying forage preservation method or supplemental protein source.

Materials and Methods

Sixteen Holstein cows in early lactation (63 d postpartum) were assigned to replicated 4 x 4 Latin squares according to calving date and covariate milk production. A 2 x 2 x 2 factorial arrangement of treatments was utilized to examine effects of dietary NDF level (26% vs 32%), forage

preservation method (alfalfa hay [H] vs. alfalfa silage [L]), and protein supplement (solvent soybean meal [S] vs. expeller soybean meal [E]). Fiber level differed across squares while the other two factors differed within squares. Periods were three weeks in length; two for adaptation and one for collection. Rations were formulated to meet fiber levels and to contain a minimum of 18% CP using dried shelled corn was the energy source. Because silage quality was very high (39% NDF, 21% CP) no S was added with L in the 32% NDF diet. Therefore, contrasts were used to test treatment combinations.

Results and Discussion

Actual ration NDF concentrations were slightly higher and CP lower than those formulated (Table 1). Milk production tended to be higher with lower diet NDF (Table 2). Milk production was significantly higher for H than L at both NDF levels but E was greater than S only at 26% NDF. Protein yield was greater for H than L and protein content was greater for S than E in the 26% NDF diets but not in the 32% diets. No fat yield or content differences were significant between any treatments nor were any significant interactions present. Protein source did not improve performance at higher NDF levels and may not affect optimum NDF level determinations when adequate CP is provided. Differences between forage preservation methods indicate possible changes in NDF composition during storage (ie greater lignin:hemicellulose ratios) which affect milk production.

Table 1. Chemical composition of diets offered at two levels of dietary NDF.[1]

	26% NDF				32% NDF			
	HS	HE	LS	LE	HS	HE	L	LE
Dry mater (%)	88.8	88.8	62.0	62.3	88.4	88.4	55.7	56.1
NEl[2] (Mcal/kg DM)	1.66	1.67	1.69	1.70	1.55	1.56	1.58	1.59
	..... % DM							
Crude protein	17.6	17.7	17.9	17.9	16.9	16.9	18.0	19.7
NDF	27.3	27.3	26.7	26.7	33.6	33.6	32.4	32.4
NDF from forage	21.3	20.7	21.8	21.5	29.8	29.6	29.9	29.4
ADF	17.6	17.2	18.4	18.1	22.9	22.7	23.7	23.7
Lignin	3.0	3.0	3.4	3.4	4.2	4.2	4.6	4.5
Forage:Concentrate	47:53	45:55	55:45	54:46	65:35	65:35	76:24	
75:25								

[1]H = Alfalfa hay L = Alfalfa silage S = Solvent soybean meal E = Expeller soybean meal
[2]Calculated from NRC (1989) and NDF content of forages.

Table 2. Production and intake parameters for cows receiving one of two levels of dietary NDF, forage, and soybean meal source.[1]

	26% Dietary NDF					Contrast		
	HS	HE	LS	LE	se	H/L	S/E	Interact
Milk (kg/d)	33.4^{b}	35.9^{a}	31.5^{c}	33.4^{b}	0.6	**	**	
4% FCM (kg/d)	30.3^{ab}	31.2^{a}	28.7^{b}	30.3^{ab}	0.6	*	*	
Fat yield (kg/d)	1.13	1.12	1.08	1.13	0.02			
Protein yield (kg/d)	1.05^{ab}	1.09^{a}	1.00^{b}	1.00^{b}	0.02	**		
Fat content (%)	3.38	3.14	3.45	3.39	0.06			
Protein content (%)	3.14^{ab}	3.03^{bc}	3.17^{a}	3.01^{c}	0.04		**	
DM intake (kg/d)	24.6	25.4	24.3	25.0	0.6			
NDF intake (%BW)	1.14	1.20	1.14	1.16	0.02			

	32% Dietary NDF					Contrast[2]		
	HS	HE	L	LE	se	S/E(H)	E(L)	H/L(E)
Milk (kg/d)	31.6^{a}	31.6^{a}	29.3^{b}	30.1^{b}	0.4			*
4% FCM (kg/d)	28.8	28.7	27.3	27.3	0.4			
Fat yield (kg/d)	1.08	1.07	1.04	1.02	0.02			
Protein yield (kg/d)	0.99^{a}	0.97^{ab}	0.92^{b}	0.93^{b}	0.02			
Fat content (%)	3.41	3.40	3.52	3.42	0.07			
Protein content (%)	3.15	3.07	3.15	3.13	0.03			
DM intake (kg/d)	24.6	23.9	23.8	23.9	0.4			
NDF intake (%BW)	1.29	1.26	1.26	1.25	0.001			

[1]H = Alfalfa hay L = Alfalfa silage S = Solvent soybean meal E = Expeller soybean meal
[2]S/E(H) = S vs E in H E(L) = E vs no E in silage H/L(E) = H vs L in E.
[abc]Means in same row with different superscripts differ (P<0.05).
* Contrast significant (P<0.05) ** Contrast significant (P<0.01).

NUTRIENT REQUIREMENTS AND FEED COSTS ASSOCIATED WITH GENETIC SELECTION FOR MILK COMPONENTS

R.G. DADO, G.E. SHOOK, and D.R. MERTENS

Introduction

Selection of sires for increased milk protein yield of their offspring has received much attention, but it is unclear what affect component selection has on net return. Selection based on net economic value increased milk production requires that variable costs, such as feed, be subtracted from market prices of each milk component. Feed costs incurred from meeting energy requirements associated with marginal component production have been determined, however, those for protein have not. Objectives of this study were to determine dietary energy and protein requirements for change in milk lactose, fat, and protein production achieved through genetic selection and to estimate marginal income over feed cost from such production.

Materials and Methods

Cows with high genetic merit were assumed to meet marginal production demands by increasing nutrient consumption. Theoretical metabolizable energy and protein requirements for each component were determined from biochemical pathways of biosynthesis. Various models were derived by changing the proportions of glucose synthesized from amino acids. Estimates were adjusted to express requirements in dietary nutrient terms. Energy and protein were adjusted according to NRC requirements and protein utilization efficiencies from nitrogen balance studies, respectively.

Shelled corn and soybean meal were used to meet nutrient requirements. Relationships between average US prices paid by producers for soybean meal and shelled corn and prices received for milk were used to estimate feed prices at current milk prices. Soybean meal and shelled corn prices were $.302 and $.127/kg DM, respectively. Milk prices (used in genetic indexes) were $1.11, $3.40, and $2.40 per kilogram of lactose, fat, and protein, respectively.

Results and Discussion

Energy and protein requirements varied with percent of glucose derived from amino acids (Table 1). Fat production required the greatest amount of energy and milk protein the greatest amount of protein, regardless of nutrient model. These nutrient models allow the calculation of nutrient requirements for milk of any composition. Feed cost was greatest and marginal return lowest with milk protein production (Table 2). At a price of $11.30 per 45.4 kg milk, the price received by producers for fat must decrease to $2.08/kg and that for protein increase to $3.92/kg, using model B, for marginal income over feed cost as a percent of gross value to be equivalent for each milk component. Appropriate emphasis for each yield trait in selection indexes for sires can be accomplished by weighting each trait according to net return above feed cost.

Table 1. Dietary energy and protein required per kilogram of marginal milk component production with different percentages of glucose derived from amino acids.

Model	Percent of glucose from amino acids	Metabolizable energy protein			Apparently digestible protein		
		Lactose	Fat	Protein	Lactose	Fat	Protein
		.....Mcal.....			.....kg.....		
A	0	5.93	13.41	7.62	0	0.01	1.31
B	5	5.98	13.42	7.60	0.09	0.06	1.21
C	10	6.03	13.43	7.58	0.17	0.11	1.12
D	15	6.09	13.43	7.56	0.22	0.14	1.00

Table 2. Feed costs and income over feed costs associated with marginal milk component production with different amounts of glucose derived from amino acids.

Model	Percent of glucose from amino acids	Feed cost[1]			Marginal income[2] over feed cost		
		Lactose	Fat	Protein	Lactose	Fat	Protein
		.....$/kg.....			.....%.....		
A	0	.178	.409	.872	84.0	88.0	63.7
B	5	.223	.435	.821	79.9	87.2	65.8
C	10	.263	.457	.778	76.4	86.5	67.7
D	15	.292	.474	.720	73.8	86.0	70.0

[1]Soybean meal (S) and shelled corn (C) were used to meet requirements. S:C price ratio = 2.38:1 with S=$0.302/kg and C=$0.127/kg.

[2]Marginal income over feed cost = $\frac{\text{(Gross value of component-feed cost of component)}}{\text{Gross value of component}}$

Milk prices: lactose=$1.11/kg; fat=$3.40/kg; protein=$2.40/kg.

LIMITATIONS TO BROMIDE DETERMINATION IN ALFALFA USING A Br^- SELECTIVE ELECTRODE

L.L. MEYERS and M.P. RUSSELLE

Introduction

Bromide has been successfully used as a tracer of NO^-_3 movement in soil-water systems and has potential for use as a tracer of nutrient uptake by roots in soil-plant systems. An important consideration in choosing a tracer is how easily it can be quantified in the system being studied. Bromide has been measured in plants and soils using several methods which traditionally have required extensive equipment, facilities, time, or expertise to complete. More recently the bromide selective electrode (BrSE) has been used to accurately determine Br^- in some plants and soils relatively quickly and easily. Interfering ions, such as Cl^- and compounds, such as proteins, in the extracts of plants or soils can lead to erroneous results with the BrSE, especially at low levels of Br^- concentration ([Br-]) such as those present in tracer studies. The objective of this research was to determine if the BrSE is suitable for use in determining low [Br^-] (1 to 10 mg/L) extracted from alfalfa.

Materials and Methods

Two extractants were tested. Dried, ground alfalfa was extracted with either 0.025 M $Al_2(SO_4)_3$ or 0.1 M $NaNO_3$. Bromide was determined using a BrSE and a method of standard additions to compensate for the complex matrix associated with plant extracts. Duplicate subsamples were also analyzed for [Br^-] by x-ray fluorescence spectrophotometry (XRFS) to verify the results. Interferences from chloride and soluble proteins were investigated. Chloride was determined in the samples and correlated to overestimation of [Br^-] by the BrSE. Extracts were heated to denature soluble proteins and then reanalyzed with the BrSE.

Results and Discussion

Extraction of alfalfa with $NaNO_3$ resulted in very unstable mV potentials compared to $Al_2(SO_4)_3$ extracts or Br^- standards in $NaNO_3$ (Figure 1) which prevented accurate determination of solution [Br^-]. Recovery of Br^- added prior to extraction was greatly overestimated in the $NaNO_3$ extracts but was accurately determined in the $Al_2(SO_4)_3$ extracts. $NaNO_3$ was therefore eliminated as a suitable extractant for alfalfa Br^- analysis by the BrSE. Verification of the BrSE data by independent analysis of the same samples using XRFS indicated that although added Br^- was accurately recovered when extracted by $Al_2(SO_4)_3$ actual alfalfa [Br^-] was overestimated by 1.3 to 14 time (Table 1).

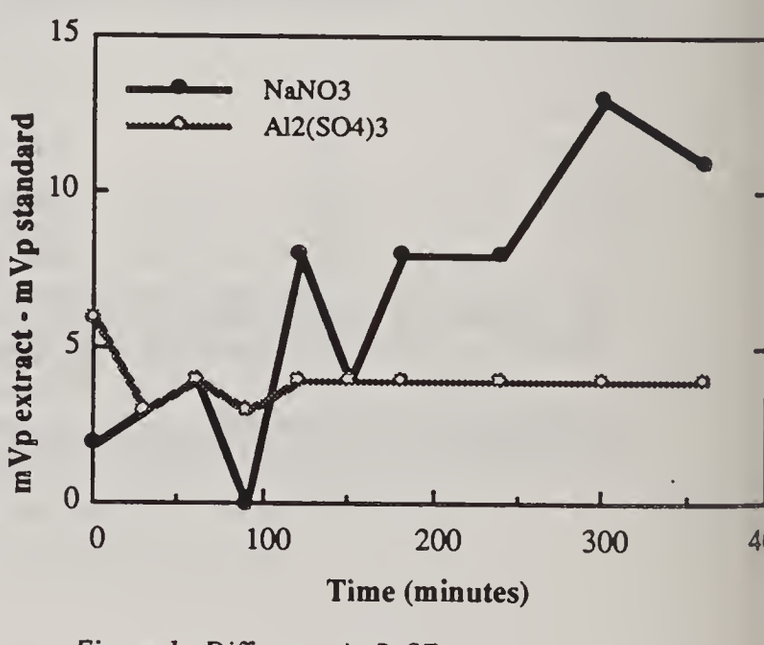


Figure 1. Difference in BrSE response (mVp) over time in two extractants.

Chloride interference will cause overestimation of [Br^-] if present in high enough concentration relative to [Br^-]. Bromide overestimation was well correlated to relative [Cl^-] and concentrations of 1 mg/L Br^- or less were overestimated even when Cl^- levels were within the recommended limit set by electrode manufacturers (Figure 2). Denaturing soluble proteins had no effect on [Br^-] overestimation by the BrSE.

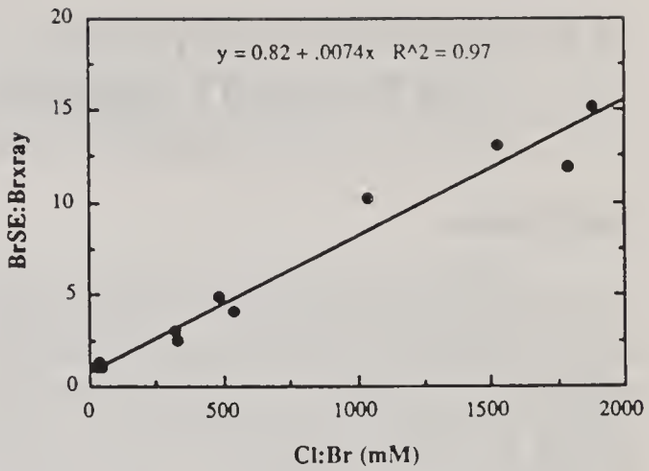


Figure 2. Relationship between [Br] determined with BrSE and XRFS at varying [Cl] to [Br] rations.

The BrSE does not appear to be suitable for determining alfalfa [Br^-] less than 2 mg/L even when [Cl^-] is within the recommended limit. The BrSE is less affected by [Cl^-] when [Br^-] is greater than 5 mg/L, yet variability of 10% was still evident at these [Br^-] levels (Table 1). Correcting for [Cl^-] may be possible, but the [Cl^-] of the sample must be known in addition to the [Br^-] and this negates the relative ease and low cost of the BrSE method.

Table 1. Apparent [Br^-] using BrSE and XRFS.

Plant Sample	[Br^-] BrSE	[Br^-] XRFS	BrSE:XRFS
	.....mg/kg.....		
1	52.4	462	1.1
2	661	497	1.3
3	58	29	2.0
4	76	22	3.5
5	100	11	9.1
6	110	10	11
7	93	8	12
8	84	6	14

ESTIMATION OF BOTANICAL COMPOSITION IN ALFALFA/RYEGRASS MIXTURES BY NEAR INFRARED SPECTROSCOPY (NIRS)

J.A. SHAFFER, G.A. JUNG, J.S. SHENK and S.M. ABRAMS

Introduction

In order to quantify the effects of climate and management practices on vegetation dynamics in pasture and hayland studies, an estimate of botanical composition is often necessary. Determination of botanical composition by hand-separation is often time consuming and expensive, limiting the number of samples that can be collected and thus limiting the scope and nature of forage studies. This study was undertaken to: 1) test the suitability of near infrared reflectance spectroscopy (NIRS) for measuring species composition in binary mixtures (alfalfa/ryegrass); 2) determine if calibration equations developed from each location were superior to equations developed from pooled samples across all locations; and 3) estimate the number of samples needed to develop adequate calibration equations.

Materials and Methods

Forage samples were collected from four different alfalfa/ryegrass trials conducted in central Pennsylvania between 1983 and 1986. All samples were harvested with hand clippers. Samples were placed in cloth bags, stored at 3°C until hand-separations were complete, and then dried at 60°C and weighed. Hand-separation involved separating the alfalfa, ryegrass and weed components from each sample. After weighing, the alfalfa and ryegrass components were remixed and ground in a Wiley mill with a 1 mm screen. Samples were frozen until analysis by NIRS.

A subset of 132 samples was randomly selected and set aside for validation of calibration equations. The remaining samples were used for calibration. Calibration equations for measurement of botanical composition (percent alfalfa) were chosen utilizing spectral and statistical analyses of the BEST program. The calibration equations were then validated by comparing actual hand-separated (reference standard) values with NIRS predicted values using coefficient of determination, bias and standard error of performance.

Several calibration equations were tested to determine if a broad-based equation derived from all four studies would be adequate, or whether calibration equations for each trial would improve precision and accuracy. Subsets of samples (approximately 5, 10, 20, 40 and 80% of those available for calibration) were also used to derive calibration equations in order to estimate the number of samples needed to obtain reliable calibration equations in alfalfa/ryegrass mixtures.

Results and Discussion

Table 1 lists the NIRS calibration and validation statistics estimated from each data set. The broad-based equations were generated from random subsets of the 1078 calibration samples collected from all years and trials. The SEP decreased rapidly with increasing number of calibration samples up to 198. Bias decreased rapidly between the 50-sample data set and the 98-sample data set, and then

remained very low. Thus, there was very good agreement between the calibration equations and the validation samples for the broad-based equations when sample size was approximately 200 or greater.

The narrow-based calibration and validation statistics for each of the four trials show a slight improvement in precision but a slight increase in bias when compared with the broad-based calibration equations of sample size greater than 200. It does not appear, therefore, that there is much advantage in taking the time and effort necessary to generate calibrations for each location and year in a multiple year and/or location study, provided that representative samples from each year and location subset are included in the overall calibration equation.

Table 1. NIRS statistics from alfalfa/ryegrass mixtures.

Source of samples	n	R2	SEC	n	r2	SEP	Bias
All trials	50	.96	6.8	132	.91	9.5	2.1
	98	.94	5.0	132	.94	8.0	-0.2
	198	.94	5.4	132	.95	6.9	-0.3
	396	.95	6.2	132	.96	6.5	0.2
	788	.96	6.1	132	.96	6.6	0.1
	1078	.95	6.5	132	.95	7.0	-0.4
Trial 1	552	.97	6.5	61	.97	5.7	0.4
Trial 2	275	.97	5.2	32	.97	6.0	-0.7
Trial 3	162	.83	6.2	21	.97	5.0	0.7
Trial 4	125	.61	7.4	18	.63	5.6	-0.4

SEC=standard error of calibration.
SEP=standard error of performance.

PROTOCOL FOR NEAR INFRARED REFLECTION CALIBRATION: MONITORING ANALYSIS RESULTS AND RECALIBRATION

J.S. SHENK, M.O. WESTERHAUS and S.M. ABRAMS

Introduction

Success of near infrared reflectance spectroscopy (NIRS) in forage analysis is dependent on the assumption that samples of interest are part of the population of samples used to generate calibration equations. NIRS analysis may disagree with reference method values when 1) the instrument is not working properly, (2) the sample population changes and is no longer represented by the calibration set, or (3) some aspect of the reference method changes, usually unknown to the analyst. None of these events must take place if NIRS analysis is to be accurate. Continual monitoring is necessary to ensure accurate results. The major cause of inaccurate results is the application of equations to populations not adequately represented in the group of samples used for calibration. The unknown samples may be from a different population than the calibration samples because of a difference in processing techniques, chemical composition outside the range of the calibration set, or application of unusual chemical or physical treatments to the samples. During routine NIRS analysis these changes are often not obvious to the technician. Thus, a system for monitoring the accuracy of an equation is necessary.

The Monitoring System

The monitoring system consists of two tests that determine the existence of (1) a significant bias and (2) a significant increase in unexplained error (SEP(C)). If either is occurring, steps should be taken to (1) add samples to the existing calibration set and recalibrate or (2) develop a new calibration for this population based on all new samples.

One sample of every 20 should be routinely set aside for reference method analysis until nine samples have been accumulated. If predictions of a group of samples appear unreasonable or the standardized H values are larger than 3.0, a random set of nine samples should be immediately selected from the group of samples exhibiting the problem. These samples should be analyzed by the reference method and the values compared with the NIRS values by the methods described here. A continuous time chart of the observed bias and unexplained error should be maintained to facilitate direction of trends.

Statistical tests are constructed for bias as estimated by observed bias, and unexplained error as estimated by SEP(C). Such tests always represent a compromise between not detecting changes that are occurring, and incorrectly identifying random fluctuation as real change. We have found that a bias greater than the standard error of calibration (SEC) and an unexplained error greater than two times the SEC are unacceptable with calibration equations based on more than 100 samples.

A full explanation of monitoring procedure, including a practical example and a discussion of the statistical basis of the procedure is presented in Supplement 2 of USDA-ARS Agriculture Handbook No. 643 (1989).

EFFECT OF IN VITRO METHODOLOGY UPON NDF DIGESTION

R.J. GRANT and D.R. MERTENS

Introduction

The Cell Wall Characterization and Utilization Work Group performed a collaborative study reported in last year's Research Summary Report which examined the differences among three current in vitro methods with respect to NDF disappearance at 0, 6, 24, 48, and 103 h post-inoculation. This research concluded that maximum extent of digestion was unaffected by in vitro technique, but could not evaluate effects of in vitro method upon rate due to small number of time points. Since rate of digestion, as well as extent, limits digestibility of a forage, the potential impact of methodology upon estimates of digestion rate must be assessed. The purpose of this research was to examine the effect of several factors which differ among the three in vitro techniques used by the Cell Wall Group upon in vitro NDF digestion. The factors included: fermentation vessel, continuous CO_2 gassing, media reduction, and use of tryptone and microminerals as nutritive additions.

Materials and Methods

Method 1 (FGRA): A .5 g sample of alfalfa or brome hay was incubated with 40 ml of Goering and Van Soest (G & VS) buffer and 10 ml of rumen fluid which had been blended for 60 s and filtered through 2 layers of cheesecloth and a 150 mesh plastic screen under CO_2 and vacuum. Buffer and samples were reduced by purging the 125 ml Erlenmeyer flasks (F) with CO_2 and injecting 2 ml of reducing solution (cysteine hydrochloride+sodium sulfide nonahydrate). The inoculum was added after solutions were reduced as indicated by change in color of resazurin indicator from purple to colorless. These flasks were kept under continuous CO_2.

Method 2 (FGRNA): This method was the same as #1 except that no nutritive additives (NA) were added to the fermentation media.

Method 3 (FGNRA): This method was the same as #1 except that no reducing solution (NR) was added to flasks prior to inoculation with rumen fluid.

Method 4 (FNGRA): A .5 g sample was incubated with 40 ml of G & VS buffer and 10 ml rumen fluid as prepared in Method 1. However, flasks were not kept under continuous CO_2 pressure (NG), but were purged with CO_2 only at time of inoculation and fitted with bunsen valves.

Method 5 (TGRA): A .4 g sample of forage was incubated with 32 ml of G & VS buffer and 8 ml rumen fluid prepared as in Method 1 except that 50 ml centrifuge tubes (T) were used instead of 125 ml Erlenmeyer flasks. All other procedures are the same as Method 1, except that only 1.6 ml of reducing solution was added to each tube.

Method 6 (TNGNRNA): This method was the same as Method 5 except that the tubes were neither purged with CO_2, kept under continual CO_2, reduced, nor had nutrients added. These tubes were fitted with bunsen valves as in method 4 and represent conditions typical of most Tilley-Terry types of in vitro systems.

Samples for all methods were incubated for 0, 4, 8, 12, 18, 24, 30, 36, 48, 72, and 96 h and analyzed for NDF remaining at each time. The 0 h time data were averaged across treatments because they had no significant effect at 0 h. The 0 h NDF value was 42.8% for alfalfa hay and 68.4% for brome hay.

Results and Discussion

Table 1 illustrates the effect of in vitro methodology upon NDF digestion averaged across both forage substrates. Differences among methods began at 8 h, were maximal at 24 h, and generally decreased thereafter through 96 h of fermentation. Comparison of methods 5 vs 6 indicates that elimination of all treatments (continuous gassing, reduction, and additives) reduces NDF digestion. Except at 24 h, fermentation vessel (method 1 vs 5) had no effect upon NDF disappearance. Therefore, fermentations in flasks were used to evaluate individual treatments. Only method 4 is consistently different from method 1 indicating that purging fermentation vessels with CO_2 at the time of inoculation and fermenting without continuous CO_2 results in less NDF digestion. Thus, continuous gassing with CO_2 appears necessary for most rapid digestion, if the buffer solutions are not bubbled with CO_2 before inoculation. The degree of NDF digestion was consistently greater for methods 2 and 3 compared to method 1, and was significant at 24 h. The significance of eliminating reducing agents and/or additives was investigated in a companion report.

Table 1. Effect of in vitro technique upon NDF digestion averaged across substrates.

Time	FGRA 1	FGRNA 2	FGNRA 3	FNGRA 4	TGRA 5	TNGNRNA 6
h	..... (% NDF)					
4	55.4	54.4	55.0	55.9	54.6	55.6
8	51.9[b]	52.6[b]	51.0[b]	54.9[a]	51.9[b]	53.5[a,b]
12	48.3[b]	47.3[b]	45.4[b]	53.6[a]	47.3[b]	50.7[a,c]
18	43.8[b]	40.9[b]	39.9[b]	49.1[a]	43.5[b]	45.7[a]
24	41.8[b]	36.9[c]	37.1[c]	44.3[a]	36.9[c]	40.6[b]
30	36.3[b]	33.6[b]	33.6[b]	40.5[a]	34.1[b]	38.9[a]
36	32.0[b]	30.8[b]	31.9[b]	39.8[a]	33.1[b]	36.6[a]
48	29.3[b]	28.7[b]	28.9[b]	33.6[a]	30.1[b]	33.2[a]
72	27.3[b]	27.9[b]	26.4[b]	28.9[a]	27.3[b]	28.8[a]
96	25.9[b]	25.2[b]	25.7[b]	28.1[a]	26.9[b]	26.9[b]

[a,b,c]Means within row differ (P<.05).

A BUFFER SYSTEM FOR pH CONTROL AND EVALUATION OF pH EFFECTS UPON FIBER DIGESTION IN VITRO

R.J. GRANT and D.R. MERTENS

Introduction

Reduction in fiber digestion typically occurs when readily available carbohydrates comprise a significant portion of ruminant diets. One mechanism by which these carbohydrates, such as starch, depress fiber digestion might be via a decline in ruminal pH when they are fed. Ruminal microbes appear to be sensitive to changes in pH, with most preferring a range of 6.5 to 6.8. Cellulolytic bacteria are generally more sensitive than amylolytic species to low pH and thus depression of fiber digestion at low pH may be due to negative effects of pH upon cellulolytic bacteria. The potential for deleterious effects of pH upon fiber digestion exists in the high-producing dairy cow whose ruminal pH may fall below 6.2 for 50 to 80% of each day. Kinetic analysis of the effect of starch upon fiber digestion at pH 6.8 has indicated that lag time increases, while digestion rate is unaffected. These results cannot explain depressions in fiber digestion observed in vivo. Effect of pH below 6.8 upon fiber digestion in batch in vitro systems has not been investigated. Therefore, the objectives of this experiment were: 1) to develop an in vitro buffering system capable of controlling pH between 5.8 and 6.8, and 2) to examine the effect of media pH upon NDF digestion after various times of incubation.

Materials and Methods

The buffering system evaluated was that of Goering and Van Soest (G & VS), adjusted to a pH of 5.8, 6.2 or 6.8 with citric or phosphoric acids. Use of phosphoric acid to adjust pH of a buffer solution has been described by Terry et al.(1969. J. Sci. Agric. 20:317). Approximate amounts of citrate added to (G & VS) buffer to obtain desired pH were: 1) pH=5.8, 960 ml buffer + 40 ml 1 M citrate, 2) pH=6.2, 982 ml buffer + 18 ml citrate. Prior to use, a 40 ml aliquot of each buffer was warmed to 39°C under CO2 and pH of the remaining media was adjusted by adding the corresponding acid before use. Samples (.5 g) of alfalfa silage and a 1:1 mixture (dry weight) of alfalfa silage:corn grain were weighed into 125 ml Erlenmeyer flasks and 40 ml of buffer and 2 ml of reducing solution were added. Flasks were incubated for 0, 12, 24, 48, and 72 h. If pH drifted downward by more than .2 pH unit, bicarbonate solution was used to adjust pH to desired level.

The experiment to evaluate the effect of media pH upon NDF digestion was replicated three times. The 3 x 2 x 6 factorial arrangement of treatments consisted of alfalfa hay, brome hay, and corn silage incubated at pH 5.8 or 6.8 for 0, 12, 24, 48, 72, or 96 h. All NDF values are reported on a blank-corrected, ash-free, dry matter basis. The inoculum source was a fistulated nonlactating cow consuming 4 kg of grain daily with alfalfa-grass hay ad libitum. The pH of inoculum at time of collection averaged 6.3. ANOVA of mean NDF remaining at each time point was accomplished using the General Linear Models procedure of SAS.

Results and Discussion

The %NDF remaining at each time point was not affected by the acid used to obtain the desired incubation pH, whether citric or phosphoric acid (Table 1). A pronounced pH effect upon NDF digestion was indicated by higher NDF residues in flasks at pH 5.8. Generally, digestion was similar

at pH of 6.2 and 6.8. Citric acid maintained pH at desired level with less additional bicarbonate solution than phosphoric acid. Since citrate exerted no apparent negative effects upon NDF digestion and resulted in the most stable pH throughout incubation, we adopted this system in further studies to examine pH effects upon NDF digestion. The pH of the in vitro system dramatically affected NDF disappearance (Table 2). With alfalfa hay, pH effects were significant at the 0 and 12 h time point. The NDF of brome hay remaining at each time point except 0 h differed with respect to pH, while corn silage differed at all times. Considering that the typical dairy cow may have ruminal pH below 6.2 for a large portion of each day, justification exists for further examination of the effect of pH upon fiber digestion kinetics.

Table 1. In vitro disappearance of NDF as influenced by pH, substrate, and acid used to adjust buffer system to desired pH.

Time (h)	pH of fermentation media 5.8	6.2	6.8
		Alfalfa silage	
0	39.0/39.0[1]	38.5/39.8	39.3/39.2[2]
12	36.8/36.5	28.2/29.6	29.1/30.5
24	29.1/28.5	25.0/25.0	24.6/24.4
48	23.2/23.6	23.3/23.6	20.6/20.8
72	22.4/22.4	21.6/21.8	20.4/19.5
		Alfalfa silage:Corn grain (1:1)	
0	25.9/26.2	25.0/26.1	27.1/28.6
12	22.3/21.7	20.6/20.2	18.9/18.4
24	16.0/16.9	13.8/14.3	13.5/14.2
48	14.7/14.6	12.1/12.9	11.4/12.0
72	14.9/13.2	10.7/11.6	10.8/10.9

[1]NDF remaining at each time when media adjusted to desired pH with phosphoric/citric acid.
[2]Media adjusted to pH 5.8 with the corresponding acid and then readjusted to 6.8 with bicarbonate solution.

Table 2. Percentage of NDF remaining at selected times of in vitro fermentation as influenced by substrate and pH of buffer.

Time	Alfalfa hay pH=5.8	6.8	Bromegrass hay 5.8	6.8	Corn silage 5.8	6.8	SEM
h	.....% NDF remaining.....						
0	43.9	42.3	68.8	67.6	36.1	35.0	.30
12	40.7[a]	34.4[b]	66.2[a]	57.3[b]	35.0[a]	30.1[b]	.77
24	29.5	28.6	51.1[a]	47.1[b]	29.6[a]	19.5[b]	1.17
48	25.1	25.4	37.1[a]	32.1[b]	21.5[a]	13.5[b]	.99
72	23.6	23.7	34.9[a]	29.6[b]	16.6[a]	13.1[b]	.43
96	24.1	23.9	32.2[a]	27.4[b]	15.7[a]	11.3[b]	.53

a,bMeans within a substrate column at each time point with unlike superscripts differ (P<.05).

FIBER DIGESTION AS INFLUENCED BY BUFFER TYPE, MEDIA REDUCTION, AND CO_2

R.J. GRANT and D.R. MERTENS

Introduction

A companion article in this year's Research Summary Report examined the effects of fermentation vessel, continuous CO_2 gassing, media reduction, and use of tryptone upon NDF digestion. In that study, the effect of reducing solution and micromineralsnutritive additives upon NDF digestion was unclear. These inconsistent observations prompted a study examining the interaction between additives and reducing solution on NDF digestion. The initial study also indicated that continuous gassing of fermentation vessels with CO_2 was critical for maximal digestion. However, apparatus for continuous gassing is cumbersome. A final study was conducted to determine if saturating the buffer solution by bubbling with CO_2 for 12 h before inoculation could replace continuous gassing. Bubbling is typically used with McDougall's buffer to adjust pH, and the evaluation of CO_2 administration and buffer type was included in the final study. Together, these experiments should suggest the best method for evaluating substrate limitations to fiber digestion kinetics.

Materials and Methods

Experiment #1: Effect of additives and reduction upon NDF digestion.

Method 1 (FGRA), Method 2 (FGRNA), and Method 3 (FGNRA) are as described in the companion report. Method 4 (FGNRNA) involved incubating a .5 g sample with Goering and Van Soest (G & VS) buffer solution and 10 ml rumen fluid without reducing solution (cysteine hydrochloride+ sodium sulfide nonahydrate) or micromineralstryptone. This design allowed comparison of all combinations to determine if addition of both reducing solution and nutrient addtives interfered with NDF digestion. Substrates included alfalfa and brome hay incubated for 0, 4, 8, 12, 18, 24, 30, 36, 48, 72, and 96 h and subsequently analyzed for NDF remaining after fermentation. The 0 h NDF values were averaged over treatments.

Experiment # 2: Effect of buffer type and CO_2 upon NDF digestion.

Method 1 (FM-CRA): A .5 g sample of alfalfa or brome hay was incubated in 125 ml Erlenmeyer flasks with 40 ml of McDougall's buffer under continuous CO_2 with additives and reducing solution described in Goering and Van Soest's procedure (USDA Handbook No. 379, 1970). 10 ml of rumen fluid were added to each flask following complete reduction as indicated by color change of resazurin from purple to colorless.

Method 2 (FVS-CRA): Same as Method 1 except that G & VS buffer was used rather than McDougall's.

Method 3 (TM-BRA): A .3 g sample was incubated in a solution of rumen fluid and McDougall's buffer in 50 ml polypropylene tubes. The buffer solution was bubbled with CO_2 for 12 h prior to inoculation and warmed to 39°C. Before inoculation, 1.2 ml of reducing solution were added to the tubes. At inoculation, micronutrients and 30 ml of a 20% solution of rumen fluid and buffer were added. The tubes were purged with CO_2 and closed with screw caps.

Method 4 (TVS-BRA): Same as Method 3 except that G & VS buffer was used rather than McDougall's.

Samples for all methods were incubated for 0, 4, 8, 12, 18, 24, 30, 36, 48, 72, and 96 h and analyzed for NDF. The 0 h NDF values were averaged over treatments.

Results and Discussion

In experiment 1 average NDF at 0 h was 43.6% for alfalfa and 68.2% for brome. Digestion was slowest when both reducing agents and nutritive additions were eliminated as indicated by higher NDF residues at 12 to 24 h of fermentation. The use of both additives and reduction together appeared to result in similar NDF digestion. However, elimination of reducing agents tended to decrease digestion during early fermentation (4 and 8 h). This agrees with the hypothesis that reducing the media prior to inoculation would minimize shock to the microbial population and reduce lag time before digestion.

Average NDF at 0 h for alfalfa and brome was 43.6 and 67.9%, respectively, in experiment 2. The most dramatic difference in this experiment was between continuous gassing of G & VS buffer in flasks compared to fermentations in sealed tubes using McDougall's buffer bubbled with CO^2. In general, continuous gassing in flasks appeared to promote more rapid NDF digestion than using sealed tubes with CO_2-saturated buffers. Within tube and flask treatments, G & VS buffer typically produced lower NDF residues signifying more rapid digestion.

In conclusion, it is recommended that an in vitro system for assessing fiber digestion kinetics should use continuous gassing and reducing agents. These treatments promote earliest initiation and most rapid digestion and provide the greatest opportunity for detecting differences in substrate related to their intrinsic properties. In vitro systems that do not maximize digestion kinetics may not detect differences in substrate due to limitations of the in vivo system. To insure that non-fiber factors do not limit fermentation, it is recommended that nutritive additives be used to insure their adequacy in promoting maximum digestion of fiber, especially with substrates low in protein and trace minerals.

Table 1. Effect of additives[1] and reduction[2] upon NDF digestion.

Time (h)	FGRA	FGRNA	FGNRA	FGNRNA
4	54.9^{a,b}	54.1^{b}	55.5^{a}	55.3^{a,b}
8	50.6^{b}	50.1^{b}	51.9^{a}	50.1^{b}
12	47.4^{b}	45.5^{b}	44.8^{b}	48.2^{a}
18	41.4^{b}	37.7^{b}	39.0^{b}	46.8^{a}
24	35.1^{b}	34.1^{b}	36.6^{b}	40.6^{a}
30	34.4^{a}	31.8^{c}	33.1^{b}	31.2^{c}
36	30.6	29.7	31.2	29.5
48	29.3	28.1	28.3	28.1
72	28.9^{a}	26.3^{b}	27.8^{b}	27.2^{b}
96	26.8	26.1	26.2	26.0

^{a,b,c}Means within rows differ (P<.05).
[1]Tryptone and Van Soest micromineral solution.
[2]Cysteine HCl and sodium sulfide.

Table 2. Effect of buffer type[1] and method of CO_2 administration[2] upon NDF digestion .

Time (h)	FM-CRA	FVS-CRA	TM-BRA	TVS-BRA
4	53.4	53.5	53.0	54.3
8	49.2[b]	48.9[b]	51.7[a]	51.4[a]
12	49.1[a]	43.8[b]	46.1[a,b]	47.8[a]
18	44.4[a]	38.4[b]	41.9[b]	42.2[b]
24	37.1[a,b]	35.2[b]	38.3[a]	37.8[a]
30	34.6[b]	33.1[b]	36.5[a]	35.7[a,b]
36	31.4[c]	33.3[b]	34.9[a]	33.0[b]
48	28.8[b]	30.0[b]	32.3[a]	29.8[b]
72	27.6[b]	27.1[b]	30.5[a]	28.7[a,b]
96	27.2	27.6	29.4	28.1

[a,b,c]Means within rows differ (P<.05).
[1]McDougall's vs Van Soest buffer.
[2]Continuous vs bubbling with CO_2.

EFFECT OF RUMINAL SAMPLING SITE ON IN VITRO FIBER DIGESTION OF FORAGES

M.A. DIEHL and S.M. ABRAMS

Introduction

Use of in vitro batch fermentation to evaluate forage quality is widespread. Within the rumen digesta, particles exist in a wide range of sizes, but seem to be stratified in relation to their location within the rumen. The density of particles tend to be inversely related to their size, so that larger particles are usually found at the top of the rumen. This coarse material conglomerates to form a mat which helps to strain larger particles, allowing smaller particles to pass through. This serves an important digestive function by: 1) retaining large particles so that they can be further digested; 2) allowing smaller particles to pass out of the rumen.

Larger particles are usually more fibrous and thus may tend to have a greater number of active cellulolytic bacteria associated with them; thus digesta from the dorsal sac of the rumen may be expected to yield inoculum of higher activity than that from other parts of the rumen. The objective of this study was to determine if a significant difference exists in the digestive capacity of inoculum obtained from the ventral versus the dorsal areas of the rumen.

Materials and Methods

Two forages were utilized, an alfalfa hay and a switchgrass/timothy hay. Inoculum was obtained from a single fistulated steer, one week apart. On the first sampling date, inoculum was obtained first from the ventral area of the rumen followed by sampling from the dorsal region. Digesta was squeezed through 7 layers of cheesecloth and transported in pre-warmed insulated containers to the laboratory. Dilution with buffer solution and inoculation of forage samples was performed in the same order as rumen sampling. On the second sampling date, the order was reversed, i.e., dorsal sampling followed by ventral sampling. Forage samples were incubated for 12 and 24 hours at 39°C, followed by neutral detergent fiber determination. Six replications of each treatment combination were used.

Results

Results are presented in Table 1. Sampling from the dorsal region resulted in a average increase of 1.0 percent in fiber digestibility compared to sampling from the ventral region ($P<.001$). The difference was more pronounced in timothy/switchgrass compared to alfalfa, probably because alfalfa fermentation was closer to its asymptotic endpoint. The difference was more pronounced after 12 hours of fermentation compared to 24 hours, probably for similar reasons. These results indicate that if maximum activity of rumen microorganisms is desired during in vitro fermentation, site of sampling should be considered, as well as other experimental variables.

Table 1. Effect of site of sampling on in vitro fiber digestion of high and low quality forages.

	Forage:	Alfalfa		Timonthy/Switchgrass	
SITE	Incubation (h):	12	24	12	24
		.....Fiber digestibility, percent.....			
Ventral sac		59.4	67.0	26.6	38.7
Dorsal sac		60.0	67.3	28.2	40.1

FUNCTIONS FOR DESCRIBING CHANGES IN DAIRY COW CHARACTERISTICS DURING LACTATION FOR USE IN DAFOSYM

D.R. MERTENS and C.A. ROTZ

Introduction

The previous Animal Submodel in the DAiry FOrage SYstem Model (DAFOSYM) used both forage and animal characteristics to develop realistic feed usage budgets within each year of multi-year simulations. Within the Animal Submodel, rations were formulated to maximize forage use while meeting target milk production levels specified by the simulator. The ration formulation system was based on the model of intake regulation proposed by Mertens (J. Anim. Sci. 64:1548, 1987). This model predicts that maximum forage usage occurs when both the energy requirement for milk

production and the fiber intake constraint of the animal are met. Thus, both fiber and energy concentration of the forage and milk production and fiber intake constraint of the animal are considered when estimating intake and formulating rations.

Body weight of lactating cows, target milk production and proportion of the herd that are first-lactation cows are inputs to DAFOSYM. In the previous Animal Submodel, the lactation cycle was divided into four stages: 0-60 days in milk, 60-150 days in milk, 151-330 days in milk and 60 days dry period. Within each of these stages a base milk production, milk fat percentage, body weight change, body weight and neutral detergent fiber (NDF) intake constraint for cows in first-lactation and second or greater lactations was established. Target milk production, body weight and body weight gain for individual simulations were adjusted proportionally based on the inputs of the simulator.

Flexibility of the Animal Submodel could be increased by making grouping strategy an input to DAFOSYM. This requires that functions be developed that describe milk production, milk fat percentage, body weight and NDF intake constraint changes over the lactation cycle so average animal characteristics can be determined for any grouping situation. The objective of this research was to develop these functions from available literature.

Materials and Methods

A modified infinite Gamma function was chosen to describe the changes in animal characteristics during the lactation cycle. This function has shape characteristics typical of animal responses, is easily fitted to data using logarithmic transformation and iterative least-squares procedures, and can generate curves that match the target inputs to DAFOSYM by changing only one parameter. This function has the form $Y = A(w+s)^{b}\exp^{[-c(w+s)]}$; where: A = the intercept, w = week of lactation, s = shift factor (in weeks), b = exponent of time, c = exponential rate of change. Parameters b and c define the shape of the curve and parameter A determines the peak and total lactational yields.

Parameters from the functions of Congleton and Everett (J. Dairy Sci. 63:109, 1980) were used to generate curves for first, second and third or greater lactations ranging in production from 4500 to 11400 kg. Regressions between function parameters and lactational yield were used to derive lactation curve parameters for a herd producing 9070 kg (20,000 lbs) milk annually which was chosen as the base production for DAFOSYM. Milk fat percentage is described by a function obtained from Williams (Ph.D Thesis, Cornell Univ., 1988). Body weight data from Davis and Hathaway (Neb. Ag. Expt. Sta. Res. Bull. No. 177, 1955) and Morgan and Davis (Neb. Ag. Expt. Sta. Res. Bull. No. 82, 1936) were used to derive parameters for the body weight function. The (A) parameters of functions were adjusted to obtain an average herd body weight of 600 kg during months 2-5 of lactation, which is the base weight used in DAFOSYM.

Data from Williams (ibid.,1988), Mertens (ibid.,1987), Dado and Mertens (J. Dairy Sci. 72:510, 1989), Cassida et al. (J. Dairy Sci. 71:381, 1988), Eastridge et al. (J. Dairy Sci. 71:2959, 1988), DePeters and Smith (J. Dairy Sci. 69:135, 1986), Woodford and Murphy (J. Dairy Sci. 71:674, 1988), Tessmann (MS Thesis, Univ. of Wis., 1988) and Hintz et al. (XVI Int. Grassld. Congr., p.813, 1989) were used to derive function parameters for NDF intake constraint. Published NDF intakes for cows in late lactation varied greatly; therefore, equation parameters were modified to be consistent with energy densities and dry matter intakes recommended by NRC (1988).

Results and Discussion

Parameters for describing changes in dairy cow characteristics during the lactation cycle are given in Table 1. These functions represent the average cow in a herd and may differ for individual cows because month of calving can alter the shape of lactation curves. Functions can be generated for different target characteristics by adjusting the (A) parameter. For example, if the target body weight (Y) is specified at any week (w) after calving, a new function can be generated that fits this condition by substituting Y, w, s, b, and c in the equation and solving for A. The situation is more complex if total lactation yield is used to derive a new function. In this case, the function must be numerically integrated (by summing the total milk yields for each week) and A is solved by iteration (initial estimates of A are adjusted until the desired total milk yield is reached).

Conclusions

These functions are not only useful for improving DAFOSYM, but also they can be used to calculate nutrient requirements, estimated dry matter intakes and recommended ration characteristics for any combination of stage of lactation and animal characteristics.

Table 1. Parameters of the modified infinite Gamma function for describing changes in dairy cow characteristics during the lactation cycle.

Trait/ Parameter	First Lactation	Second+ Lactation
Milk Yield (kg/d)/		
s	0	0
A	24.1	34.0
b	.178	.221
c	-.0210	-.0340
Milk Fat (%)/		
s	0	0
A	5.0	5.0
b	-.24	-.24
c	.016	.016
Body Weight (kg)/		
s	12/7	11/7
A	567	690
b	-.0730	-.0803
c	.0087	.0072
NDF Intake Constraint (%BW/d)/		
s	6/7	21/7
A	.564	.388
b	.360	.588
c	-.0186	-.0277

DAFOSYM: AN AID FOR STRATEGIC PLANNING OF DAIRY FARMS

C.A. ROTZ, D.R. MERTENS, D.R. BUCKMASTER and J.R. BLACK

Introduction

The dairy forage system involves complex interrelationships between crop production, harvest, storage, and utilization by the dairy herd. The many factors involved make it difficult to determine the costs and benefits of implementing various management and technological changes on dairy farms. Changes which benefit one part of the system must be examined carefully to ensure that they do not cause equal or greater negative impact on other parts of the system. Conditions may also vary considerably from year to year. A system which performs well one year, may or may not perform well over the weather patterns of other years.

DAFOSYM is a computer simulation model of the dairy-forage system developed to evaluate the economic consequences of management decisions and technology from a total system perspective over a wide variety of weather conditions. The model was originally developed as a research tool. Several new technologies under development have been extensively evaluated to determine their value to the farmer. The model has also been used to compare management strategies such as the type and size of machinery and storage structures, methods of harvest, crop mixes, farm size, etc. An objective of this years work on DAFOSYM was to complete a user-friendly, well documented model for distribution to other potential users.

Materials and Methods

DAFOSYM integrates crop growth, harvest, storage, feeding, and animal utilization. Corn and alfalfa growth are simulated using historical weather data. Alfalfa harvest includes mowing, field curing, raking, baling, and chopping; losses and quality changes are modeled as functions of the mechanical treatment, field curing time and weather. Storage losses and quality changes are functions of the type of storage and the crop moisture content. Stored feeds are supplemented with soybean meal, distillers grain, corn grain, and purchased hay to provide balanced diets for a given herd. Costs of crop production, machinery, storage facilities, labor, and purchased feeds are subtracted from the income from milk and excess feed sales to determine the return above feed costs. A multiple year simulation evaluates the long-term benefit and risk of a technology or strategy on a representative dairy farm.

Results and Discussion

DAFOSYM is a useful tool for teaching or demonstrating the inter- relationship of the many parts of the dairy forage system. The model also may be useful to farmers and farm consultants as they make strategic decisions that impact the dairy farm. DAFOSYM is written in ANSII Fortran 77 and compiled for use on personal computers with a DOS operating system. A user- friendly menu driver allows rapid change of major model parameters. The model requires about 1.0 megabyte of disk space for storage and 512 kilobytes of RAM for execution. The model is most conveniently used on a personal computer with a fixed disk and execution time is greatly reduced when a math coprocessor is used.

A reference manual and user guide were written to document the current version of DAFOSYM. The reference manual describes the many algorithms, major assumptions and equations used to model the dairy forage system. The manual includes a description of the verification or validation of major portions of the model. The user's guide describes the procedure for using the model including descriptions of the various input menus, procedures for modifying the input files and an interpretation of the results obtained. A copy of the model, a users guide, and a reference manual can be obtained by request to C.A. Rotz.

U.S. DAIRY FORAGE RESEARCH CENTER
ANNUAL DAIRY OPERATIONS REPORT, FEBRUARY 1990

L.L. STROZINSKI

The research center herd count presently stands at 530 (260 cows and 270 herd replacements). We are currently milking 215 cows which are yielding an average of 65.4 pounds of milk per day. Our DHIA rolling herd average stands at 16,511 pounds of milk, 606 pounds of fat and 507 pounds of protein. These averages have dropped during the past year. Although numerous factors have contributed to this decrease, I feel the most influential factors have been research pressure, inadequate staffing and feeding program changes resulting from the drought. Hopefully these averages have bottomed out and will take an upward trend in 1990. The reproductive performance of the herd continues to be excellent. Average days from calving to conception is presently 99 days yielding a calving interval of 12.6 months. Our heifers' average age at first calving is 24 months. Our herd is young in that 34 percent of the milking herd are first calf heifers and the average age of the milking herd is 48 months. Our goal is to increase this average age toward 60 months.

During the past year increased emphasis in the breeding program has been placed on protein production, stature and strength. Continued emphasis has been placed on milk production and cheese yield dollars. Sire selection is made from the top 400 U.S. sires with the aid of two computer sire selection programs (Maxbull from Virginia Tech and the Bull-search from the Holstein Association). We have also been using a computerized mating service from 21st Century Genetics.

Research usage of the herd continues to be high throughout the year as it should be. Presently 57 percent of the milking herd are on research trials. Significant progress has been made in the areas of planning and coordination to distribute the research workload more evenly across the available workforce.

The dairy unit has operated during a large portion of the past year with vacant full time positions. Temporary help, students and assistance from the field operation have kept the unit going. Action to fill the vacancies is moving slowly through the appropriate channels and I hope to fill all positions this spring.

The new double eight Boumatic herringbone milking parlor which was installed a year ago has worked out quite well. Cow throughput has increased. Cow I.D. and milk weights are now collected and transferred to the farm computer automatically. Milk weight data is then transferred to the campus computer daily.

The manure separator designed and constructed by Dr. Koegel and his staff has been installed in the manure processing room and has been functioning well for the past six months. The new unit has a higher throughput than the old machine and is considerably less costly to operate in terms of man hours, down time and repairs.

The center continues to host many visitors from around the world throughout the year. One June 20, 1989 the Forage Center cooperatively hosted the Wisconsin State Forage Council field day which was attended by approximately 800 people. An area Wisconsin Agricultural and Life Sciences Alumni Association picnic was also held at the farm on that day.

U.S. DAIRY FORAGE RESEARCH CENTER
ANNUAL FIELD OPERATIONS REPORT, FEBRUARY 1990

B.C. VENUTO

The 1989 cropping season was slightly below normal but much improved over 1988. Rains were timely all season long and just adequate to maintain decent crop growth. We have not replenished subsoil moisture and are very dry at lower depths. With this winter's (1989-90) poor snow cover to date and deep frost, we will need above normal rainfall this spring and summer of 1990 if we are to replenish subsoil moisture.

Our leased acreage was increased by approximately 90 acres in 1989. Most of this area was used for additional corn production. The 1989 corn trial was moved into this area and averaged 125 bushels per acre. These yields were well below previous years (with the exception of 1988).

Alfalfa stands were hurt somewhat by winterkill. Low areas, areas with short stubble and wheel track areas were killed due to ice sheeting. The winter was particularly kind to weevils. The weevils were not however kind to the alfalfa. Stands seeded in 1988 suffered most weevil damage.

Alfalfa yields were fair overall with second crop considerably below average and third crop somewhat above average. Quality was quite good overall.

In addition to an increase of acreage in 1989 we also expanded our hay storage by adding an 80 foot extension to our existing 50' x 200' open front shed. This will allow us to use our storage building in Badger Ordinance for equipment and research material storage and enable us to store all our bedding needs closer to the barns.

Another priority for 1990 will be the planning of a chemical storage and handling facility. This is a timely need. We should not ignore our responsibility to lead by example in this area.

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